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The Plant Disease Reporter is issued as a service to plant pathologists throughout the United States. It contains reports, summaries, observations, and comments submitted voluntarily by qualified observers. These reports often are in the form of suggestions, queries, and opinions, frequently purely tentative, offered for consideration or discussion rather than as matters of established fact. In accepting and publishing this material the Crops Research Division serves merely as an informational clearing house. It does not assume responsibility for the subject matter.

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ACCEPTANCE OF MANUSCRIPTS

The increase in the volume of pertinent material offered for publication in the Plant Disease Reporter has made it necessary to limit the subject matter and the length of articles accepted. The subject matter should emphasize new things in plant pathology, such as new records of disease occurrence, serious outbreaks and epidemics, conditions affecting development of plant diseases, techniques of investigation including instrumentation, new discoveries in control including new materials and their evaluation. Manuscripts will be limited to 15 double-spaced typed pages. Insofar as possible, material should be presented as graphs rather than tables. Papers cannot be accepted for publication that report routine control experiments, reviews, bibliographies without annotation, results of routine surveys, mere summaries or lists of plant diseases. By following this procedure we hope to continue publishing all articles promptly.

Paul R. Miller

Manuscripts for and correspondence about this publication should be sent to:

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CONTROL OF LEAF RUST OF WHEAT WITH INORGANIC NICKEL[1]

F. R. Forsyth and B. Peturson[2]

Summary

Nickel salts were applied for rust control on field plots of Thatcher wheat at Winnipeg in 1957-58. Nickel chloride and nickel nitrate hexahydrates applied at rates of from 6/10 to 1 pound per acre in 66 gallons of water eradicated wheat leaf rust and caused increases in yield varying from 8 to 21 percent. Available evidence indicates that the 1 pound per acre rate of application is the most suitable. Spraying at 7 or 8 day intervals during the period after a trace of rust is present in the crop is suggested.

Nickel has been shown to be effective in greenhouse experiments as an eradicant of rye leaf rust, Puccinia rubigo-vera (DC) Wint. f. sp. secalis Wint., wheat leaf rust Puccinia rubigo-vera (DC) Wint., and wheat stem rust Puccinia graminis f. sp. tritici Eriks. and Henn. (1, 2, 3). The effectiveness of certain nickel salt amine complexes as rust control chemicals in the field has also been reported (5). However, inorganic nickel was found to have less persistence as a protective fungicide than the organic forms and was not considered likely to be successful in practical control of rust (1, 2). Tests with simulated rain under greenhouse conditions showed that protective properties of the simple nickel salts were easily lost (1, 2).

Results at Winnipeg in 1957 and 1958 in field tests, reported in this paper, have convinced us that the inorganic salts of nickel, particularly the chloride and nitrate, can be used to control leaf rust and presumably also stem rust of wheat in the field.

METHODS

The wheat variety Thatcher was used both in 1957 and 1958 in the field plot tests. It was grown in rows 1 foot apart in plots 4 feet by 18 feet with four buffer rows between test plots. There was one row of winter wheat between each plot and the buffer rows beside it. In 1957 there were four replicates and in 1958 six replicate plots for each check and fungicidal treatment. The locations of the plots within a block were chosen at random, each block containing one plot of each treatment and one check plot.

Infection by stem and leaf rust was due entirely to naturally occurring inoculum in 1957 but in 1958 a leaf rust epiphytotic was artificially induced in the field plots by dusting a mixture of leaf rust urediospores and talc (1:100) onto the rows of winter wheat. Both in 1957 and 1958 stem rust was too small a proportion of the total rust on the wheat in the plots to be more than a minor factor in the experiments. In 1958 stem rust never exceeded a trace amount in the test plots.

The fungicides were applied with knapsack sprayers of 1 gallon capacity. A weighed amount of fungicide was applied to each plot in 500 ml of water by adjusting the rate of walking so that all liquid was applied during one trip along the rows. This amount of water per plot is the equivalent of 66 gallons per acre.

The criteria used to evaluate the effectiveness of the chemicals under test were: the percentage of rust at the final reading by use of the modified Cobb's scale (4); the weight of 1000 kernels; the yield in bushels per acre; and the weight in pounds per bushel.

In 1957 we included nickel nitrate treatments in our field plots of the U. S. D. A. Cooperative Rust Fungicide Test. Only the results of the check, treated check and nickel-treated plots are presented here. The treated check was included to give a measure of the yield of the variety under test in the virtual absence of rust. Lately, zineb has been used for this purpose on a protective schedule but sulfur has been equally effective in past years. In 1958 an experiment was carried out to compare the effectiveness of 1, 2 and 3 pounds per acre of nickel chloride

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[1] Contribution No. 15 from the Canada Department of Agriculture Research Laboratory, Winnipeg, Manitoba.

[2] Respectively, Plant Physiologist and Plant Pathologist, Plant Pathology Section.

in the control of rust on Thatcher wheat on protective (5 day interval) and eradicative (10 day) spraying schedules. The plants were growing rapidly at the time of first application of nickel salts. This may have contributed to the phytotoxicity noted with the 2 and 3 pounds per acre rates. Consequently only the modified schedule shown in Table 1 for experiment 1958A was completed.

Experiment 1958B was a portion of the 1958 U. S. D. A. Cooperative Rust Fungicide Test at Winnipeg and only the part dealing with inorganic nickel is presented here.

The data on yield, bushel weight and 1000 kernel weight were statistically analysed by means of the analysis of variance method.

RESULTS

The results of the field plot evaluation of inorganic nickel as a fungicide in three separate experiments are presented in Table 1. It is obvious that the use of several applications of zineb as a protective fungicide, either by tank-mix through the use of nabam plus zinc sulfate or by wettable powder, resulted in higher yields per acre and greater 1000 kernel weights than did the use of inorganic nickel. However, most of the increases due to the application of inorganic nickel were statistically significant and were achieved at a low cost of chemical and a low number of applications.

The increases in yield per acre and 1000 kernel weight resulting from the use of nickel on a protective schedule (5 day interval) were greater than those caused by nickel on an eradicative (10 day) schedule. Although the schedules were such that protective action would be expected in the short interval, there would also be eradicative action. Since spraying the wheat crop every 5 days would not be feasible, we attach most significance to the results obtained with nickel applied on the 10 day schedule. Perhaps a 7 or 8 day interval between sprayings would be better than the 10 day interval used in these experiments.

The eradicative nature of the rust control with nickel was apparent because of the presence of dead or arrested infections on plants sprayed on a 10 day schedule. A dark ring of discolored tissue formed around the infection site of the pustule and further development of the pustule was arrested. The fact that readings such as 25-30 percent rust (all dead or arrested) were found when a 10 day period between sprayings was used indicated that in the 10 day interval infections begin and reach a visible stage before they are killed by the next application of chemical.

Phytotoxicity was not found with the 1 pound per acre applications. The 2 and 3 pound per acre rates caused necrosis averaging 10-15 and 25-40 percent respectively of the leaf area, due to the phytotoxic effects of a single application of nickel chloride. The one application of nickel chloride at the rate of 3 pounds per acre reduced 1000 kernel weights and yields per acre of plots so treated to less than the corresponding values for check plots.

DISCUSSION AND CONCLUSIONS

The inorganic salts of nickel appear to fulfill most of the requirements sought in a chemical for rust control. Most important, they are therapeutic and are thus independent of weather conditions during the spraying season. The cost of the chemical and of its application should be low if 2, 4-D ground equipment is used. The wheat grower can wait until rust is present in the crop before he commences spraying. This has two advantages, first that he will not be too late in commencing spraying, a real possibility when protective fungicides are used, and second that the total length of the spraying period is shortened. The long storage life of the nickel salts is also an important consideration in the control of the wheat rusts because rust epidemics are of sporadic occurrence, and yet the supply of chemical for rust control must be readily available on short notice when required.

Evidence to date indicates that two or three applications at 7 or 8 day intervals, of nickel chloride or nitrate hexahydrate at 1 pound per acre in 50 gallons of water, would give reasonable control of wheat leaf rust and presumably of wheat stem rust in the field.

The following points remain to be elucidated:

1. The amount of residual or accumulated nickel in the grain due to spraying for rust control.

2. The tolerance level of nickel allowed by government regulation on cereal grain and its processed products.

3. The effect on subsequent crops of residual nickel in various types of soil.

Table 1. Result of applications of nickel salts and zineb for rust control on field plots of Thatcher wheat at Winnipeg, 1957-58.

Treatment	Dates of application	Pounds/acre	Number of applications	Percentage leaf rust July 12, 1957 Aug. 1, 1958	Percentage leaf rust Aug. 6, 1957 Aug. 19, 1958	Yield of check in bu./acre and diff. from check	Average 1000 kernel weight of check and diff. from check	Weight per bushel (pounds)
1957								
Check			0	Trace on 95% plants	80 on top leaf	20.8	20.64	55.6
Nabam + $ZnSO_4$	July 13, 20, 29 Aug. 3, 9	1.5 as resultant zineb	5	Trace on 95% plants	12 on top leaf	+12.2	+6.54	+4.6
Nickel nitrate	July 19, 29, Aug. 8	0.6, 0.6 and 1.1	3	Trace on 95% plants	16 on top leaf	+3.5	+1.30	+2.4
					L.S.D. 5%	3.28	1.25	1.48
					L.S.D. 1%	4.44	1.68	1.99
1958-A								
Check			0	5-10 on top leaves	90 on top leaf	61.5	25.95	61.4
Zineb	July 3, 12, 17, 18, 25 Aug. 1, 8, 15	2	8	Trace on top leaves	5-10 all infections alive	+17.9	+6.52	+1.1
Nickel chloride	July 18, 23, 28 Aug. 1, 7, 12, 18	1	7	Trace on top leaves	5-10 all dead	+13.2	+5.98	+0.9
Nickel chloride	July 18, 28, Aug. 7	1	3	Trace on top leaves	15-25 all dead	+5.3	+2.18	+0.9
Nickel chloride	July 18, Aug. 1, 12	2	3	1-5 on top leaves	25-35 all dead or arrested	+0.8	+2.22	+0.6
Nickel chloride	July 18	3	1	1 on top leaves	60-70 all alive	-10.7	-1.37	-0.4
					L.S.D. 5%	6.7	2.43	0.82
					L.S.D. 1%	9.2	3.31	1.12
1958-B								
Check				5-10 on top leaves	85-90 all alive	53.3	25.84	62.5
Zineb	July 21, 31	2	8	Trace on top leaves	10 all alive	+19.7	+5.96	+0.1
Nickel nitrate	July 21, 31	0.6	2	3-5 on top leaves	60-75 all alive	+8.6	+1.86	+0.8
					L.S.D. 5%	7.7	1.09	0.74
					L.S.D. 1%	10.3	2.56	0.99

4. The effect on germination of the seed harvested from treated plants.
5. The effect on baking quality of the flour.

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CANADA DEPARTMENT OF AGRICULTURE RESEARCH LABORATORY, WINNIPEG, MANITOBA

RESIDUAL ACTIVITY OF THREE FUNGICIDES APPLIED TO THE SOIL FOR BUNT CONTROL[1]

Laurence H. Purdy[2]

Summary

The residual duration of three fungicides applied to soil were determined, using percentages of common bunt as a measure of the effectiveness of each fungicide. Anticarie at 2 1/2 and 5 pounds per acre was ineffective after 6 months, while applications of 20 pounds per acre were effective for 6 months but for less than 12 months. Panogen 15 and Terraclor were not effective after 6 months at the 3 rates tested.

The soil-inhabiting nature of dwarf bunt (Tilletia contraversa Kuehn) of winter wheat in the Pacific Northwest constitutes a major obstacle to the control of this disease. Perennial soil infestation precludes successful control by crop rotation, and host resistance is successful only within the limits of the inherent resistance of adapted varieties to the prevailing races. The use of effective chemical control measures, therefore, has become increasingly important.

A possibility for chemical control was indicated in 1957[3] when it was shown that 40 percent hexachlorobenzene (HCB) applied to the soil at the rate of 10 pounds per acre was effective against this disease. Even though excellent control was obtained in the 1957 experiments, the necessary application rate of 10 pounds per acre may pose economic considerations detrimental to the general adoption of soil treatment with HCB as a control measure. Any benefits, as measured by the control of dwarf bunt, that might be derived from the residual effects on succeeding wheat crops would reduce the cost of soil treatment. Since the duration of activity of HCB applied to the soil had not been determined, the study reported here was initiated to determine whether significant control of bunt infection from soil-borne spores could be realized by the residual carryover of fungicides from one season to the next.

MATERIALS AND METHODS

The fungicides tested were Anticarie (40 percent hexachlorobenzene), Terraclor (40 percent pentachloronitrobenzene), and Panogen 15 (2.2 percent cyano [methylmercury] guanidine). Anticarie (HCB) and Terraclor (PCNB) were applied to the soil of individual plots at the rates of 2 1/2, 5, and 10 pounds per acre in a water suspension of 300 ml per plot. Panogen 15 was applied in water dilutions of 1:10,000; 1:1,000; and 1:10 by using 300 ml of each dilution per plot. The treatments were randomized and replicated three times at each treatment date.

Bottomless wooden frames, 24 x 36 inches, made of 2 x 6 inch stock, were placed on the soil surface and filled to within 2 inches of the top of each frame with field soil, a Palouse silt loam. The wooden framework defined the limits of each plot and held the soil in place. Ninety-nine plots were established by this method.

Because high percentages of infection are difficult to obtain from artificial inoculations with dwarf bunt, common bunt (T. caries (DC.) Tul.) was used to determine the effectiveness of the soil treatments. Each plot was inoculated with a suspension of bunt spores applied at the rate of 1/2 gram of spores per square foot of soil surface.

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[1] Cooperative investigations of the United States Department of Agriculture, Agricultural Research Service, Crops Research Division, and the Agricultural Experiment Stations of Idaho, Oregon, and Washington. Scientific Paper No. 1787, Washington Agricultural Experiment Stations, Pullman, Washington.

[2] Pathologist, United States Department of Agriculture, Agricultural Research Service, Crops Research Division, Pullman, Washington.

[3] Purdy, L. H. 1957. Differential response of dwarf bunt to seed and soil surface treatment with hexachlorobenzene. Plant Disease Reptr. 41: 916-918.

The fungicides and spores applied to the soil surface of each plot were worked into the soil to a depth of 2 inches prior to planting.

One test was made with Red Bobs spring wheat and all others with Orin winter wheat. The seed of both varieties was clean and free of bunt spores.

The soil of all plots was inoculated, treated, and the seed was sown on the dates indicated in the following tabulation:

Plot number	Date inoculated	Date treated	Date sown
1-33	April 24, 1956	May 3, 1956	May 3, 1956
	November 2, 1956		November 5, 1956
	November 6, 1957		November 8, 1957
34-66	November 2, 1956	November 5, 1956	November 5, 1956
	November 6, 1957		November 8, 1957
67-99	November 6, 1957	November 8, 1957	November 8, 1957

Each plot was treated once with a test fungicide. As shown in the above tabulation, the soil of plots 1-33 was inoculated, treated, and seeded in the spring of 1956. These same plots were reinoculated and seeded in the fall of 1956 but were not treated again. Similarly, the soil of plots 34-66 was inoculated, treated, and seeded in the fall of 1956. These plots were reinoculated and seeded again in the fall of 1957 but were not treated again. The soil in plots 67-99 was inoculated, treated, and seeded in the fall of 1957 and thus served as a check against plots 1-66, which were reinoculated and seeded, but not retreated, at the same time.

Bunt percentages, based on the total number of heads per plot, were the measure of effectiveness of the various treatments.

RESULTS

The results, as average percentages of bunt, are presented in Table 1. Only 7 percent bunt developed in the check plots in the first seeding made in plots 1-33. Good control was obtained with all three rates of application of Anticarie and Terraclor. Panogen 15 at the dilution of 1:10 provided good control but was phytotoxic and resulted in a slightly reduced stand. The other two rates of Panogen 15 provided only fair or no control.

In the second seeding, following reinoculation, in plots 1-33, 57 percent bunt developed in the check plots. Only Anticarie, at 20 pounds per acre, showed any residual effect, reducing the bunt incidence to 3 percent. In plots 34-66, which were inoculated and treated, good control was obtained with Anticarie and Terraclor at all rates of application and with Panogen 15 at the 1:10 dilution rate. The best control was obtained with Anticarie and Terraclor at the two higher rates of application (Table 1).

Table 1. Average percentages of common bunt in experimental plots to determine the residual duration of three fungicides applied to the soil.

Fungicide	Pounds per acre	Percent bunt in plots: 1-33 Treated May 1956	34-66 Treated November 1956	67-99 Treated November 1957
Inoculated and seeded April and May 1956:				
Anticarie	2 1/2	1		
	5	T[a]		
	20	0		
Terraclor	2 1/2	1		
	5	T		
	20	0		
Panogen 15	1:10, 000	2		
	1:1; 000	6		
	1:10	0		
Check - untreated		7		

Table 1 (continued)

Fungicide	Pounds per acre	Percent bunt in plots: 1-33 Treated May 1956	34-66 Treated November 1956	67-99 Treated November 1957
Inoculated and seeded November 1956:				
Anticarie	2 1/2	60	6	
	5	63	4	
	20	3	T	
Terraclor	2 1/2	50	18	
	5	53	1	
	20	60	T	
Panogen 15	1:10,000	67	70	
	1:1,000	50	68	
	1:10	51	12	
Check - untreated		57	68	
Inoculated and seeded November 1957:				
Anticarie	2 1/2	12	4	10
	5	12	7	T
	20	4	8	T
Terraclor	2 1/2	7	6	5
	5	12	7	5
	20	9	11	0
Panogen	1:10,000	9	10	15
	1:1,000	11	12	15
	1:10	13	8	3
Check-untreated		11	8	17

[a] Trace, less than 1/2 percent bunt.

In the third seeding, the bunt incidence was relatively low in the untreated check plots, ranging from 8 to 17 percent (Table 1). In plots 67-99, treated at the time of planting, good control was obtained with Anticarie at the 5- and 20-pound rates, with Terraclor at the 20-pound rate, and with Panogen 15 at the 1:10 dilution rate. These same treatments had no residual effects, as indicated by the lack of significant reduction of the bunt incidence in plots 1-66 (Table 1).

CONCLUSIONS

The good control obtained from soil applications of Anticarie (HCB) at 5 and 20 pounds and Terraclor (PCNB) at 20 pounds per acre at the time of seeding shows that the use of these chemicals and rates could be practical and economic measures for the control of bunt where resistant varieties have failed to provide adequate protection. The results show clearly that the residual life of Anticarie (HCB) at 2 1/2 and 5 pounds per acre is less than 6 months, while applications of 20 pounds per acre showed activity for at least 6 months but less than 12 months. It is also shown that the residual effectiveness of Terraclor (PCNB) and Panogen 15 applied to soil is less than 6 months at the three rates tested. Although the application of HCB to the soil provides control of dwarf bunt, these results indicate that there is no significant residual benefit to succeeding crops.

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TWO PREVIOUSLY UNREPORTED FUNGI ON CEREALS IN MONTANA[1]

E. L. Sharp

Abstract

Surveys were made to determine the fungi causing root and foot rots of cereals in Montana. Cercosporella herpotrichoides and Cephalosporium gramineum, previously unreported in Montana, were found in various wheat-growing areas of the State. Other fungi commonly isolated from diseased roots and crowns were Helminthosporium sativum, Fusarium roseum f. cerealis, Ophiobolus graminis, Hendersonia crastophila, Gloeosporium bolleyi and Pythium spp.

Fungi causing root and foot rots of cereal crops constitute a serious disease problem in Montana. During the growing seasons of 1957 and 1958 surveys were made to determine the cause and distribution of cereal foot and root rot diseases.

Diseased roots and crowns of the cereal (wheat, barley, and oats) were collected from various grain-growing areas of the State and brought into the laboratory for isolation work. The diseased areas were sectioned into 1-cm lengths, washed for several hours in tap water, surface sterilized with 70 percent alcohol and 20 percent clorox solution, washed in two separate sterile water blanks and then plated on either corn meal or potato-dextrose agar.

Cercosporella herpotrichoides Fron., and Cephalosporium gramineum Nisikado and Ikata, previously unreported in Montana, were found in various wheat-growing areas of the State. C. herpotrichoides was found in Prairie, Cascade, Teton, and Pondera counties and C. gramineum was isolated from wheat plants collected in Gallatin, Cascade, and Big Horn counties. A fairly wide distribution of these organisms indicates that they have not been recently introduced into the State. More intensive surveys would probably reveal an even wider distribution of these pathogenic fungi.

The strawbreaker fungus, C. herpotrichoides, caused breaking of culms in localized lower areas of fields. Cercosporella was in a late stage of development on the hosts when isolated and was often accompanied by Hendersonia crastophila Sacc., as reported by Sprague (4). However, isolations from fallen foot-rotted grain most often resulted in Helminthosporium sativum P.K.B., Fusarium roseum f. cerealis Snyder & Hansen, and H. crastophila rather than the strawbreaker fungus. Cercosporella was found on most of the common winter wheats in the State and also on Compana barley.

The organism causing the Cephalosporium stripe disease, Cephalosporium gramineum, was isolated this past growing season from wheat grown in three counties of the State. It was first isolated in this laboratory from a wheat sample collected by Purdy[2] in a field near the regional smut plots at Bozeman. Cephalosporium was isolated from Yogo, Cheyenne and Itana wheat. Isolations were made from plants showing striping of culms. The diseased plants often exhibited considerable darkening around the nodes. Sterile heads, similar to take-all infected plants, were abundant. The Cephalosporium stripe disease has been known in Japan for some time (3), but was first reported by Bruehl in 1956 to occur in this country in the State of Washington (1). It has since been found in New York State (5). Montana isolates of the fungus grown on potato-dextrose agar were similar in cultural characteristics and spore measurements to those described by Bruehl (2) for Washington isolates. Most cultures were whitish to grayish in color, but several produced yellowish sectors. The rate of mycelial growth and sporulation was greatly enhanced by the addition of peptone (1 percent) to potato-dextrose agar. Pathogenicity of the fungus was proven by inoculation of Baart wheat. Greenhouse and field experiments have been initiated to test the resistance of wheat varieties grown in Montana.

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[1]Contribution from Montana State College, Agricultural Experiment Station, Bozeman, Montana. Paper No. 444 Journal Series.

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Cercosporella herpotrichoides and Cephalosporium gramineum do not cause the most serious cereal root and foot rot diseases in the State, but they are important problems in localized areas and are a constant threat to grain growing. The most prevalent root and foot rot fungus of cereals in Montana is Helminthosporium sativum. It is widely distributed, causes considerable damage each year, and is often accompanied by Fusarium roseum f. cerealis. The latter fungus also causes considerable damage to oats in several western counties of the State and has been particularly bad in that area the past 2 years. The take-all fungus, Ophiobolus graminis Sacc., also caused considerable crop damage to wheat in various areas in the State and has been particularly destructive in Cascade county. In several fields it caused irregular patches where all plants were somewhat dwarfed and usually contained sterile white heads. Perithecia were observed rarely under field conditions, but did develop readily on plants grown in the greenhouse in soil collected from the diseased areas. Gloeosporium bolleyi Sprague was associated at one time or another with most of the other fungi and Pythium spp. were frequently found in the earlier isolations.

Experiments are being conducted to test the efficacy of various cultural practices, residues and fungicides for use in control of cereal root and foot rot diseases.

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MONTANA STATE COLLEGE, AGRICULTURAL EXPERIMENT STATION, BOZEMAN, MONTANA

A METHOD OF EVALUATING THE REACTION OF BARLEY SEEDLINGS TO INFECTION WITH SEPTORIA PASSERINII SACC.[1]

Adam F. Koble, Glenn A. Peterson, and R. G. Timian[2]

Abstract

The importance of developing a method for the evaluation of the reaction of barley seedlings to infection with Septoria passerinii Sacc. is discussed. A procedure for testing seedlings in the greenhouse and the results of several experiments are presented.

Disease development in barley seedlings susceptible to S. passerinii was best when plants were incubated at high humidity for 5 days at a temperature of 21° C. Significant differences in reaction of barley seedlings to cultures of varying ages and to inoculums of varying spore concentrations of the organism were obtained. The best differentiation in reaction type between resistant and susceptible varieties was observed 16 days following inoculation.

It is believed that the procedure described may be useful in greenhouse tests for evaluating varieties and hybrid material for reaction to S. passerinii.

Septoria leaf blotch caused by Septoria passerinii Sacc. recently has become an increasingly important disease on six-row spring barley in the upper midwestern area of the United States. This disease has been an important factor in reducing yields and kernel plumpness. The symptoms of Septoria leaf blotch on barley were described by Weber (3) in 1923.

Several years of testing under field conditions showed that several barley varieties are resistant to S. passerinii; however, none are of suitable malting quality and few are of desirable agronomic type. The incorporation of Septoria leaf blotch resistance in a barley of desirable malting quality necessitated the development of techniques to obtain a uniform degree of infection and a reproducible system for rating disease reaction. Such a procedure would be used to screen segregating populations for resistance and to study the mode of inheritance of disease reaction.

This paper presents a method for testing seedlings in the greenhouse whereby it is possible to obtain a uniform symptom expression of Septoria leaf blotch on barley varieties. Some of the factors affecting greenhouse testing were length of high-humidity incubation, temperature, spore concentration in the inoculum, age of the Septoria culture, and cultural differences among Septoria isolates.

In 1957 Green and Dickson (2) reported that plants inoculated by spraying with a water suspension of conidia or pycnidiospores were infected satisfactorily when incubated 48 hours in a moist atmosphere at about 20° C. The writers were unable to obtain suitable infection by use of Green and Dickson's procedure. Methods to attain complete and uniform infection were, therefore, investigated.

MATERIALS AND METHODS

Barley plants were grown in 3 1/2-inch clay pots at 21° C in the greenhouse. The plants were inoculated at the 3-leaf stage and grown under the same conditions unless stated otherwise. Inoculum was from isolates of S. passerinii collected at Langdon, Park River, and Fargo, North Dakota, and grown on potato-dextrose agar. The desired spore concentration was obtained by dilution with distilled water.

Twenty-two isolates of the fungus (designated A to V) were obtained from infected barley

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leaves from the three locations mentioned. These isolates were maintained in test tubes, and transfers were made periodically to provide cultures of the desired age for inoculation. Cultures used for inoculation were usually about 10 days old.

Inoculum was prepared by blending spore-bearing mycelial mats with distilled water and straining through a single thickness of cheesecloth. Gelatin as a sticking agent was added to the inoculum to give a 0.5 percent solution and a small amount of Tween 80 was added as a spreader. The inoculum was applied with a Venturi atomizer with as little air pressure as possible to obtain a fine mist. Plants were moistened with the inoculum until just prior to runoff and kept in chambers at or near 100% relative humidity and 21° C. Two days after inoculation the plants were moved to the greenhouse and placed in humidity chambers for an additional 3 days unless stated otherwise.

Disease readings were taken 14, 16, and 18 days following inoculation. Only the 16-day readings are presented, as symptoms were fully developed and variety differential greatest after 16 days. The amount of infection present was recorded on a scale of 0 to 5 (no symptoms to severe symptoms) which was based on both number and size of lesions. Plants receiving readings of 3 and below were classified as resistant and those receiving readings higher than 3 as susceptible. Classification of individual plants appeared possible in these experiments, but readings represent an average of all plants in a single pot because of the uniformity of symptoms on plants within varieties given the same treatments. At least three replications per treatment were used in these experiments. Each replication consisted of 10 to 12 barley seedlings per pot.

RESULTS AND DISCUSSION

Preliminary tests showed the necessity of a post-inoculation period of high humidity. Experiments were conducted to determine the optimum length of incubation at high humidity to obtain maximum symptom expression on the susceptible barley variety C.I. 10002. These tests were run at temperatures of 20° and 24° C; however, since a significant difference between temperatures was not obtained at the 5% level, the disease indices given in Table 1 are the average of the combined readings at the two temperatures. Duration periods of high humidity ranging from 36 hours to 9 days were used. The data in Table 1 show that treatments of 72 hours or less of high-humidity incubation were insufficient to obtain satisfactory disease development; however, disease development was satisfactory when high humidity was maintained for longer periods. Significant differences at the 1% level were obtained with the analysis of variance among treatments receiving additional high humidity and those receiving no additional high humidity. The application of Duncan's multiple range test (1) showed that the difference between no additional high humidity and either alternate or continuous days was significant at the 1% level. There was no significant difference at the 5% level between disease indices of treatments receiving additional high-humidity incubation on continuous or on alternate days. Resistant readings were obtained on all plants receiving no additional high-humidity incubation. Significant differences at the 5% level were not obtained among disease indices for 36 to 72 hours of initial high humidity; however, a trend of increasing values is indicated.

Additional data were obtained from tests in which the duration of high-humidity incubation varied from 2 to 14 days. High humidity for 5 consecutive days was required to obtain maximum symptom expression on the susceptible variety C.I. 10002. High-humidity incubation beyond 5 days gave no significant increase in symptom expression. The resistant variety C.I. 4439 used in these tests gave a resistant reading of 2 or lower with all treatments. This shows that an incubation period of from 5 to 14 days could be used to differentiate between resistant and susceptible varieties, such as C.I. 4439 and C.I. 10002, respectively.

A series of experiments were conducted to learn more about S. passerinii. Four isolates of S. passerinii which varied in color, rate of growth, and rate of sporulation were used in one of these experiments. The variations in pathogenicity of cultures of these isolates when 3, 6, 12, or 24 days old, on the resistant variety C.I. 4439 and the susceptible variety C.I. 10002, are shown in Table 2. The analysis of variance indicated no significant difference at the 5% level in pathogenicity among the four isolates. Significant differences among the different-aged cultures were obtained, however. The variety C.I. 4439 showed a resistant reaction with all the treatments. The variety C.I. 10002 gave a susceptible reaction with 3- and 6-day-old cultures of all isolates and with the 12-day-old cultures of isolates I and R, but a resistant reaction with the 12-day-old cultures of isolates J and D and with the 24-day-old cultures of all isolates. This shows a loss in pathogenicity of isolates with the age of the culture. The difference in reaction type between varieties was significant at the 1% level.

Table 1. The effect of duration of high-humidity incubation on the expression of Septoria leaf blotch symptoms on the susceptible barley variety C.I. 10002 following an initial high-humidity period of 36 to 72 hours at 21° C.

Initial high-humidity period (hours)	Additional high-humidity incubation		
	None	Alternate days for a total of 9 days	Continuous for a total of 9 days
36	1.33[a]	3.83[a]	4.16[a]
48	1.33	4.00	4.00
60	2.50	4.00	4.33
72	3.00	4.50	4.50

[a]Combined average readings of three replications at two temperatures based on a scale of 0-5 (no symptoms to severe symptoms).

Table 2. Disease reactions of a susceptible and a resistant variety of barley inoculated with different-aged cultures of four isolates of S. passerinii[a].

Isolate	Age of culture (days)	Resistant C.I. 4439	Susceptible C.I. 10002
I	3	3.00[b]	5.00[b]
	6	2.20	4.80
	12	2.20	4.80
	24	2.20	2.60
R	3	2.20	3.60
	6	2.80	4.40
	12	2.40	3.60
	24	1.80	1.80
J	3	3.00	5.00
	6	2.40	4.80
	12	1.80	2.60
	24	1.40	1.00
D	3	2.00	3.60
	6	2.20	4.00
	12	2.00	3.00
	24	1.80	2.20

[a]Spore concentration of each inoculum was adjusted to 225,000 spores/ml except that the 3-day-old J isolate contained 170,000 spores/ml.
[b]Average readings of five replications based on a scale of 0-5 (no symptoms to severe symptoms).

The four isolates just listed and three additional ones were tested for pathogenicity in a second experiment. Susceptible varieties expressed resistant and susceptible reactions to the various isolates, even though spore concentration and age of culture were constant. Further testing may show that the isolates can be differentiated on barley varieties.

The effects of spore concentration of the inoculum on barley seedlings of a resistant and a susceptible variety are shown in Table 3. Significant differences at the 1% level were obtained between varieties and among spore concentrations. Resistant readings were obtained on C.I. 4439 regardless of the spore concentration used; however, a concentration of between 56,000 and 112,000 spores/ml was necessary to obtain a susceptible reaction on C. I. 10002. Although similar readings were recorded on plants inoculated with 112,000 and 225,000 spores/

Table 3. Reaction of two barley varieties inoculated with various spore concentrations from the R isolate of S. passerinii.

Spores/ml	Resistant C.I. 4439	Susceptible C.I. 10002
14,000	1.00[a]	1.67[a]
28,000	1.33	1.67
56,000	1.33	2.33
112,000	3.00	4.00
225,000	3.00	4.33

[a]Average readings of three replications based on a scale of 0-5 (no symptoms to severe symptoms).

ml as shown in Table 3, additional tests showed that more consistent results were obtained when 225,000 spores/ml were used. Use of a spore concentration of 225,000 spores/ml with an optimum temperature and humidity period resulted in a disease index of 4 or higher for the susceptible variety C.I. 10002. When a similar procedure was used, resistant varieties did not receive a reading higher than 3.

Pycnidial development was observed on the susceptible varieties but not on the resistant varieties in the tests described. Varietal disease readings obtained in the greenhouse on seedling plants appear closely and positively correlated with those taken in the field in 1955 to 1957. It is believed that in greenhouse tests the procedure described may be useful for evaluating varieties and hybrid material for reaction to S. passerinii.

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INCIDENCE OF BROWN-SPOT OF CORN IN MISSISSIPPI IN 1957 AND ESTIMATIONS OF ITS EFFECT ON YIELD[1]

James W. Broyles[2]

Abstract

The chief importance of brown-spot of corn (Physoderma maydis Miyabe) is its ability to reduce yield and cause severe lodging in certain hybrids. Losses as high as 67 percent in grain and 100 percent lodging were obtained by artificial inoculation, indicating the pathogenic potentialities of the fungus. Damage caused by various levels of infestation under artificial and natural epiphytotic conditions was determined. Losses of 50 percent in individual inoculated plots were not uncommon, and losses in fields infested under natural conditions ranged up to 25 percent. A 250-field sample examined over the State of Mississippi in 1957 had an estimated reduction in yield of 1.9 percent.

Brown-spot (Physoderma maydis Miyabe) can reduce yield and cause much lodging of certain corn hybrids. Environmental conditions affect epidemiology profoundly, thus limiting damage to corn grown in the hot, humid southeastern regions of the United States. In Mississippi corn planting may extend over a period of 3 or 4 months and weather conditions are likely to be favorable for serious development of the disease in some of the plantings. Brown-spot has been the most important disease of corn in the State in recent years. In the past four seasons the disease was found in almost every field of corn examined; however, its severity in different areas varied greatly from year to year. Early surveys in Mississippi indicated losses as high as 6 to 10 percent in certain areas[3]. These reports, however, were merely guesses, and no precise information on brown-spot damage other than general distribution had been obtained until the present work was undertaken.

Brown-spot reduces corn yields, but the damage may be inconspicuous and often is underestimated. The causal fungus flourishes under much the same conditions as does corn, and the greatest losses occur when yields would normally be high. The most outstanding effect is weakening of the stalk and the resultant breakage, often attributed to other causes. Differences exist among very susceptible hybrids in the amount of lodging sustained, and certain susceptible hybrids appear to stand up well even when attacked by the fungus. Some hybrids lodge at an earlier stage than others, thus increasing losses. It is estimated that brown-spot is responsible for the greater part of the stalk lodging of corn caused by disease in Mississippi. It appears likely that earlier estimates of losses may have been too low and that the importance of the disease was not fully recognized. Consequently, experiments were begun to determine the damage caused by brown-spot as well as the pathogenic potentialities of the causal fungus.

In 1955 the effects of different levels of infestation, obtained by artificial inoculation, on lodging and yield were tested on a very susceptible single cross (NC7 x C.I. 21). Plots of 40 plants each having the desired level of infestation (light, moderate, and heavy) along with chemically treated, disease-free checks were selected. Incidence and severity of infection, percentage of lodging, and yield were determined on 6 to 10 plots representing each level. Results of this test are given in Table 1.

Differences in the amount of lodging and grain yield were very large between different levels of infestation. Heavily infected plants in this test, however, suffered considerably more damage than did plants generally considered to be heavily infected under natural conditions. The single cross NC7 x C.I. 21 is extremely susceptible to lodging under heavy infection, and there was much lodging in the test. As high as 100 percent stalk lodging occurred in some plots (Figure 1).

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[2]Pathologist, Crops Research Division, Agricultural Research Service, United States Department of Agriculture.

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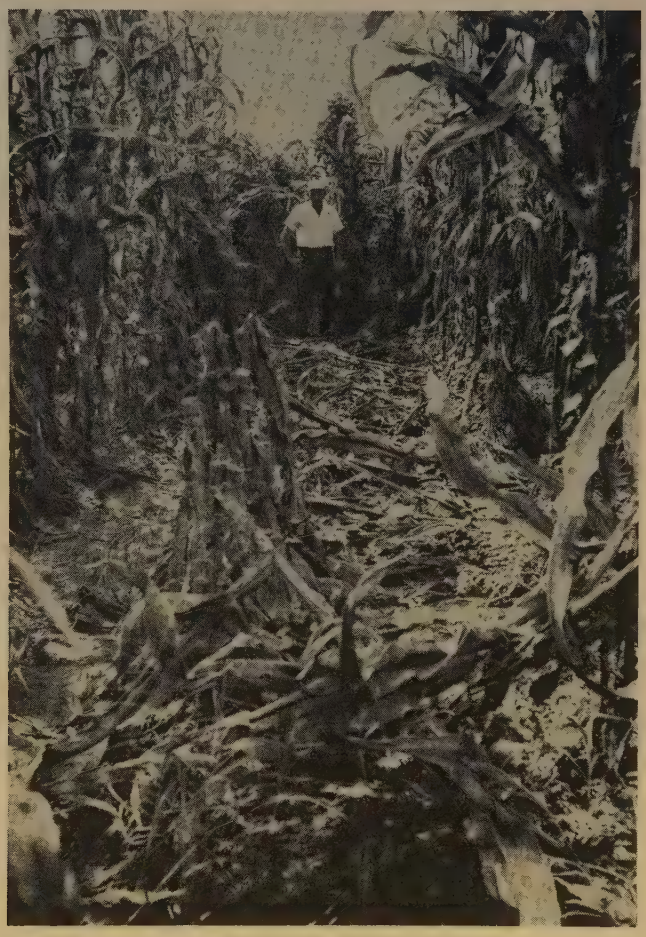

FIGURE 1. Experimental plot of NC7 x C.I. 21 corn with 100 percent stalk lodging caused by brown-spot. Adjoining rows were chemically treated to control the disease.

The average loss in grain for heavily infested plots was 43.5 percent, but some plots sustained considerably more damage than others. Comparative data from single adjacent chemically treated disease-free and heavily infested plots are shown in Table 2. Yields should have been comparable in these plots except for differences caused by brown-spot. The effects of P. maydis on yield, lodging, ear number, kernels per ear, and grain size are very evident.

In 1957 losses sustained by commercial hybrids due to brown-spot were determined under conditions of natural infestation. The effects of severity of infection on yield, and size and weight and number of first and second ears were determined in fields at State College and at Carthage, Mississippi. Each of four uniform blocks marked off in the test field represented single replications. Shortly after the corn had tasselled the levels of infection that could be used in the particular test were determined, and each plant accurately representing one of these levels was tagged. One hundred plants were selected for each level in each replication. This technique insured uniformly infected samples representing each level of infection and provided disease-free checks. All plants of all samples were intermingled within each replication; therefore, the potential yielding capacity of each sample should have been the same except for the effects of the disease. At State College only two levels of infection, moderate and disease-free, were used; at Carthage three levels, moderate, light, and disease-free, were used. Samples used to determine average field losses were taken at random and included all degrees of infection. Results of these tests are shown in Table 3.

At both light and moderate levels of infection there was approximately the same amount of stalk infection; in addition, there was considerable leaf-blade infection at the moderate level. Light infection reduced yields 11 percent as compared with 14 percent for moderate infection. This bears out the observation that leaf-blade infection causes much less damage than does stalk infection.

Table 1. Lodging and reduction in yield of corn in relation to severity of brown-spot infection.

Severity of infection	Infection (percent)	Lodging (percent)	Yield (bushels per acre)	Reduction in yield (percent)
Disease-free	0	0	81.2	----
Light	71	14	77.5	4.6
Moderate	95	49	62.5	23.0
Heavy	97	80	45.9	43.5

Table 2. Comparative data on chemically treated disease-free corn plot and adjacent plot heavily infested with brown-spot.

Severity of infection and reduction[a]	Infection (percent)	Lodging (percent)	Yield per acre (bushels)	Ears per plant (number)	Seeds per ear (number)	Weight of 100 seed (grams)
Disease-free	0	0	94.9	1.9	543	34
Heavily infested	100	100	31.7	1.2	395	25
Reduction			67 percent	37 percent	27 percent	26 percent

[a]Reduction caused by infection based on difference between infected and disease-free plots.

Table 3. Effect of brown-spot infection on corn yield and size and number of ears at two locations in Mississippi in 1957.

Location and severity of infection	Yield per acre		Weight of first ear		Weight of second ear		Second ears per plant	
	Actual (bu)	Reduction[a] (percent)	Actual (gm)	Reduction[a] (percent)	Actual (gm)	Reduction[a] (percent)	Actual (no.)	Reduction[a] (percent)
State College:								
None	73.3		161		107		.92	
Moderate	62.9	-14	152	-6	78	-27	.82	-11
Average[b]	70.9	- 3	166	+3	99	- 8	.92	0
Carthage:								
None	91.5		274		169		.79	
Light	81.5	-11	261	-3	130	-23	.69	-13
Moderate	79.0	-14	267	-1	112	-34	.59	-25
Average[b]	81.9	-11	274	0	112	-34	.67	-15

[a]Based on difference between plots with no infection and those of different levels.
[b]Samples taken at random to indicate average for the field.

Reduction in yield in predominantly two-eared hybrids, such as were used in these tests, appears to be manifested in the smaller size and number of second ears. At low levels of infection the weight of second ears is first affected, and numbers are reduced only after damage is increased somewhat. Weight of first ears is not appreciably affected except at fairly high levels of infection, when most second ears do not develop.

A survey of the incidence of brown-spot in Mississippi was made in 1957; a total of 256 fields was examined and the plants were grouped into five rough classes based on severity of infection. Fields were rated and grouped according to the percentage of plants in each infection class, and estimates of reduction in yield were made on the basis of the information gained in the foregoing experiments and in others. To determine total percent loss for each field, the incidence in each class was multiplied by the estimated loss caused by that class and these figures for all classes were added. These data are presented in Table 4.

Table 4. Estimated reduction in yield caused by brown-spot in a 256-field sample of commercial corn in Mississippi in 1957.

Infection rating of field	Percent of fields	Percent plants in indicated infection class					Percent loss per field	Total percent loss for sample
		none	very light	light	moderate	heavy		
Trace	74.6	95	5	0	0	0	0.1	0.07
Very light	14.8	50	20	25	5	0	4.0	0.59
Light	5.1	25	15	30	25	5	9.0	0.46
Moderate	3.5	10	5	35	40	10	12.0	0.42
Heavy	1.6	0	5	20	50	25	17.0	0.27
Very heavy	0.4	0	1	5	20	74	25.0	0.10
Total								1.91

Although nearly 75 percent of the fields were essentially free from brown-spot, with only about 5 percent of the plants very lightly infected, the estimated reduction in yield was 1.9 percent. These estimates were based on the effects of the disease on only certain very susceptible hybrids. Other hybrids and varieties with the same amount of infection might not suffer such great losses, but severity of infection as measured by size, number, and location of lesions is a good indication of the amount of damage. Further work is being done to test the validity of these estimates.

MISSISSIPPI AGRICULTURAL EXPERIMENT STATION, STATE COLLEGE, MISSISSIPPI, AND CROPS RESEARCH DIVISION, AGRICULTURAL RESEARCH SERVICE, UNITED STATES DEPARTMENT OF AGRICULTURE

SURVEY OF FUNGI ASSOCIATED WITH WHITE CLOVER STOLONS[1]

Norman E. McGlohon[2]

Within the past few years, as the livestock industry has increased in the Southeastern United States, increasing interest has been given the disease losses in forage crops. At Clemson a program was initiated to investigate those diseases damaging to white clover, with primary emphasis devoted to those diseases which might contribute to the lack of summer persistence. Preliminary investigations revealed that various fungi were associated with white clover plants as this summer and early fall degeneration was occurring. Thus, as a preliminary to this study, it was necessary to find out which specific fungi are associated with white clover stolons during the summer and the relative prevalence of these fungi.

This survey was conducted in two phases. The first phase was designed to find out the relative prevalence of the various fungi during the summer months. The second phase was to find out the distribution of these fungi over the State of South Carolina. Each week 50 stolons were taken from the same field in the Clemson area in Oconee County and 50 stolons were taken from at least one other county in South Carolina, sampling a different county each week for an 8-week period. On July 14, Transylvania County, North Carolina, which borders South Carolina, was sampled.

Each week beginning June 9, 1958 white clover stolons were selected at random from plants which may or may not have appeared to be diseased. Fifty stolons came from the Clemson field and 50 from one or two other locations in the State or in one case from a border county of North Carolina. The stolons were taken directly to the laboratory where they were thoroughly washed. Five nodes were removed from each stolon and dipped for 10 seconds in 95 percent ethyl alcohol. The nodes were then surface sterilized for 90 seconds in a sodium hypochlorite (Clorox) solution containing four parts water and one part Clorox and plated on acidified potato-dextrose agar. After about 5 days the fungi were identified by microscopic examination.

Table 1. Fungi associated with white clover stolons. Clemson, South Carolina 1958.

Date	Nodes tested	Sterile	Fusarium	Colleto-trichum	Rhizoc-tonia	Macro-phomina	Curvu-laria	Tricho-derma
6/9	250	11	193	6	9	26	3	4
6/17	250	18	159	13	14	38	3	5
6/23	250	3	160	16	47	8	13	3
6/30	250	18	186	1	119	3	5	2
7/7	250	4	192	6	66	1	37	11
7/14	250	3	202	15	29	11	21	29
7/21	250	6	210	8	4	8	34	124
7/28	250	6	208	2	36	1	6	12

The results of this survey are listed in Tables 1 and 2. Species of Fusarium, Colletotrichum, Rhizoctonia, Macrophomina, Curvularia, and Trichoderma were present in practically every location checked. Fusarium spp. were found associated with about 70 percent (Table 3) of all stolons taken from the Clemson field throughout the entire sampling period. They were also the leading species found in every county except Florence. Rhizoctonia was also observed each week and in all locations. At Clemson this fungus increased rapidly until June 30, after which time it decreased sharply until July 21. Curvularia was abundant in Greenville County on June 30 in a river bottom pasture area which had been extensively grazed. Trichoderma seemed to be present more often in areas where the stolons were shaded by dense top growth. At Clemson there was a definite negative correlation between the population of Trichoderma and that of Rhizoctonia, as is shown graphically in Figure 1.

[1] Technical Contribution No. 303 of the South Carolina Agricultural Experiment Station, Clemson Agricultural College, Clemson, South Carolina.

[2] Research Assistant.

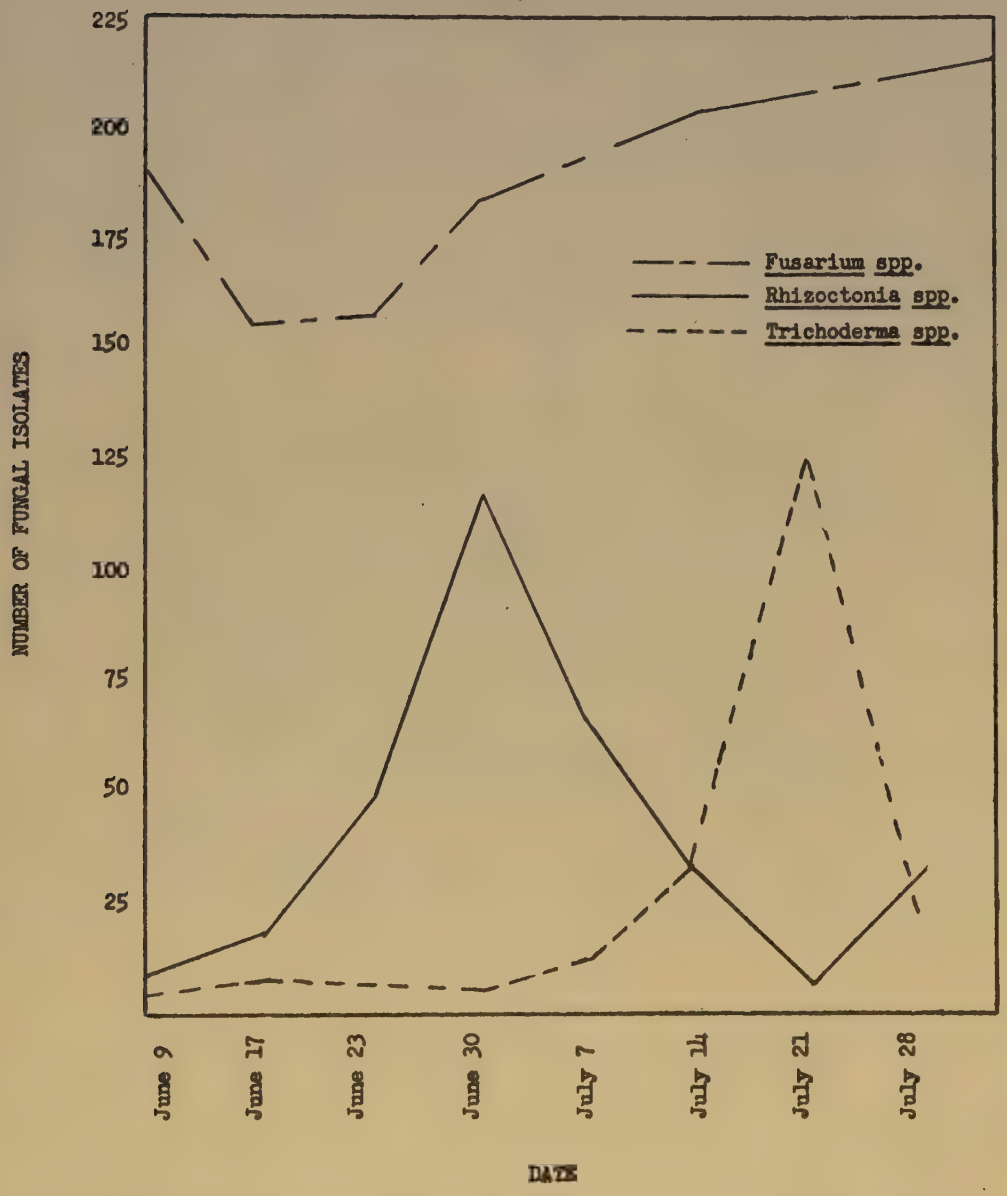

FIGURE 1. Fungi associated with white clover stolons at Clemson, South Carolina. June and July, 1958.

Table 2. Fungi associated with white clover stolons in South Carolina and one adjacent county in North Carolina in the summer of 1958.

County	Date	Nodes tested	Sterile	Fusarium	Colleto-trichum	Rhizoc-tonia	Macro-phomina	Curvu-laria	Tricho-derma
Barnwell	6/9	250	42	172	12	5	15	4	2
Spartanburg	6/17	250	8	176	2	60	3	3	1
Laurens	6/17	250	6	136	6	94	24	1	4
Greenville	6/23	250	0	200	43	17	1	146	2
Chester	6/30	250	1	167	10	41	4	2	53
Transylvania, N. C.	7/7	250	26	140	26	43	4	12	11
Hampton	7/21	250	26	172	18	62	2	4	42
Dorchester	7/21	250	28	176	6	30	1	6	58
Florence	7/22	250	42	50	34	54	14	0	56

Table 3. Summary of fungi isolated from 4250 clover nodes. Clemson, South Carolina 1958.

Fungus	Number of isolates	Percent
Fusarium	2899	70
Rhizoctonia	730	17
Trichoderma	419	9
Curvularia	300	7
Colletotrichum	224	5
Macrophomina	164	4

In some instances the fungi may have been playing the role of saprophytes rather than destructive parasites. However, if we use the prevalence of each fungus as an indication of its potential importance, we might consider Fusarium and Rhizoctonia as warranting major emphasis in the research program.

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ABNORMALITIES OF GRASS ROOTS AND THEIR RELATIONSHIP TO ROOT KNOT NEMATODES

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In 1954 Ivanoff[1] reported the occurrence of hyperplastic abnormalities on the roots of oats, barley, wheat, and other grasses. He postulated that these were induced either by ectoparasitic nematodes or by the mass action of root knot nematodes. The observations reported below present evidence supporting the latter hypothesis as well as a further record of the occurrence of these abnormalities.

In testing various common weedy grasses for susceptibility to Meloidogyne incognita var. acrita, abnormalities or galls were noted that did not contain adult nematode females. The most distinct reaction was the localized swelling of roots to about twice the normal diameter (Fig. 1).

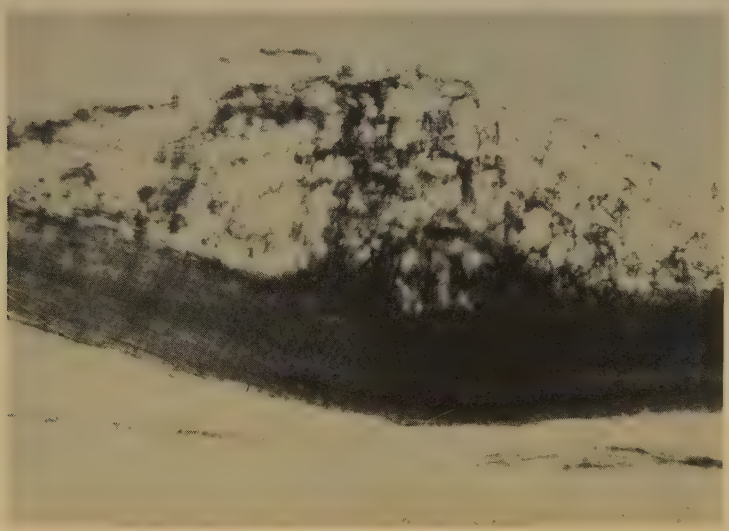

FIGURE 1. Hyperplastic abnormalities on the roots of sandbur (Cenchrus pauciflorus).

Galls were located on any part of the root system and roots with galls appeared to be stunted. In some cases the secondary roots were stunted and proliferated, but this was rare. The following grass species were observed to have these root abnormalities: Cenchrus pauciflorus (sandbur), Setaria lutescens (yellow foxtail), and Aegilops squarrosa. In the same test wheat, oats, corn, barley, and other grasses supported reproduction of nematodes on their roots, in contrast to the report of Ivanoff. The reason for this difference may be the use of different varieties of these plants or the species of Meloidogyne may have been different.

Following these observations, a test was conducted to see whether the high population of root knot nematodes present in the soil was responsible for these abnormalities. A large amount of washed galled tomato roots was added to sterile soil, and seed of Aegilops squarrosa was planted. Controls were planted without the galled roots. Two days later after the seed had sprouted, samples were taken to determine if the nematodes had invaded the roots. Upon staining the roots with hot lactophenol cotton blue for 1 minute, abundant invasion of nematodes was found in most of the roots (Fig. 2). At this time the roots appeared normal.

Two or 3 weeks later abnormalities appeared on the roots. Control plants had normal roots. After the abnormalities had appeared, the roots were examined for nematodes, with negative results. Ivanoff[1] was also unable to find any nematodes in the root abnormalities.

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[1] Ivanoff, S. S. 1954. Hyperplastic abnormalities of roots of oats and other cereals and grasses suspected to be caused by nematodes. Plant Disease Reptr. Suppl. 227: 84-85.

FIGURE 2.
Nematodes in the roots of Aegilops squarrosa 2 days after germination of host.

From this evidence, it would appear that high populations of root knot nematodes can induce abnormalities and galls on roots of grasses which will not support reproduction of the nematodes. After inducing the galls, the nematodes either die in the roots or migrate out of the roots.

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RELATION OF ROOT-KNOT NEMATODES AND IRRIGATION WATER TO THE INCIDENCE AND DISSEMINATION OF BACTERIAL WILT OF BEAN[1]

M. L. Schuster[2]

Summary

The relation between infection by root-knot nematodes and the bacterial wilt organism (Corynebacterium flaccumfaciens (Hedges) Dows. var aurantiacum Schuster and Christiansen) in a susceptible bean variety (University of Idaho No. 59) was studied in greenhouse experiments. Treatments with and without root wounding were made with and without bacteria. Plants grown in soil infested with Meloidogyne incognita and the bacteria showed a large amount of wilting in one experiment but in another comparable experiment a substantial decrease in wilting and increase in root necrosis resulted. M. hapla did not affect the rate of wilting of the beans in comparable tests. This species of nematode in combination with bacteria decreased the yield of seed and stunted the bean plants to a greater degree than did the other treatments. Under certain circumstances it appeared that the nematode, M. incognita, provided wounds through which the bacteria could enter. In one series of experiments where severe galling resulted, bacterial infection was inhibited, due presumably to a morphological or physiological effect on the host. Mechanical root wounding showed no direct effect on the plants in absence of bacteria as evidenced by yield and wilting data.

Seed infection under the conditions of the experiment is not common since infected seed were found in only one treatment in which bacteria were added to wounded roots.

Dissemination of the bacteria by irrigation water was demonstrated in greenhouse tests but negative results were obtained in field trials, indicating that spread in the field may not occur in this manner.

The data indicate that nematodes may be involved in providing portals of entry for the wilt organism under field conditions. In conjunction with cultivation injury and natural wounding the bacterial wilt organism should be capable of infecting bean plants under field conditions.

Research on the effect of nematodes on the incidence of bacterial and fungus diseases has been reviewed recently by Stewart and Schindler (4). Certain bacterial wilt organisms apparently require wounding of tissues before invasion can occur. The orange-colored strain of the bean wilt organism, Corynebacterium flaccumfaciens var. aurantiacum, is not capable of infecting the leaves of bean (Phaseolus vulgaris) through the stomata (3). This is also true for C. flaccumfaciens (5). Presumably these bacteria invade plant tissue through wounds. The relation of nematodes to bacterial wilts has recently been demonstrated for the carnation wilt, caused by Pseudomonas caryophylli Burkh. (4), and for Granville wilt of tobacco induced by P. solanacearum E. F. Sm. (2).

Nematodes have been suspected of providing wound-entry points for bean wilt bacteria. Greenhouse experiments designed to study the influence of mechanical root injury and contaminated irrigation water on the spread and development of bean wilt disclosed a possible influence of the root lesion nematode also. Furthermore, the bacterial organism proved capable of inducing wilt of beans when applied to nonsterilized soil but not sterilized soil, indicating a possible organism complex (3). These observations motivated the investigation of the effect of plant parasitic nematodes on the bacterial wilt of bean.

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[1] Published with the approval of the Director as Paper No. 924, Journal Series, Nebraska Agricultural Experiment Station.

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MATERIALS AND METHODS

In a greenhouse experiment to determine the relation between bacterial wilt, mechanical wounding of roots, and dissemination by irrigation water a raised bench was planted to four rows of U.I. 59 variety of Great Northern beans. Two rows represented controls and two rows were infested by applying C. flaccumfaciens var. aurantiacum to the irrigation water at the upper end of each row. To each row 48-hour-old cultures from 12 Petri dishes containing 2 percent potato-dextrose agar were applied in irrigation water 16 days after planting. Mechanical wounding of roots was accomplished by drawing a sharp knife alongside the bean rows about 2 inches from the stems and to a depth of about 4 inches. Root wounding preceded bacterial application in each experiment. In all four rows, roots in three 4-foot sections were injured and those in three similar lengths of row remained uninjured. Mechanical wounding of roots was made on only one side of the plant. Watering during the course of the experiment was by furrow irrigation. The number of wilted plants was recorded periodically. Seed from the plants was harvested and checked for visible bacterial infection; isolations were made from stems of wilted plants to determine the presence of the orange-colored pathogen.

Field tests to demonstrate the spread of wilt bacteria by irrigation water relationship in the greenhouse were conducted at the Scotts Bluff Experiment Station, Mitchell, Nebraska for the years 1954, 1955, and 1957. In these experiments an arrangement similar to that in the greenhouse tests was used, except on a larger scale. Each of three replications consisted of two adjacent rows instead of one, as was set up in the greenhouse. The length of rows was 96 feet. Each row consisted of three sections of beans with roots wounded and three not wounded.

Effect of nematodes on bacterial wilts in the greenhouse was studied by using plants grown in steamed 6-inch clay pots filled with autoclaved soil consisting of one part sand and one part compost. The pots were sunk in sphagnum moss in wooden flats to prevent rapid changes in moisture content of the potted soil. Three pots per flat represented a replication. Seed of U.I. 59, a Great Northern bean variety, was germinated in ragdolls. Three-day-old seedlings were transplanted in the potted soil, three per pot.

Nematodes for inoculation were obtained from cultures maintained on host plants growing in sterilized soil. The nematode inoculum was applied to the soil about 1 inch below the transplanted seedlings. In Experiment 1, the nematode inoculum consisted of 10 egg sacs of Meloidogyne incognita Chitwood and in Experiment 2, galled carrot roots. In the latter experiment the infected roots washed free of soil were chopped, mixed, and separated into 9-gram aliquots before inoculation.

The bacterial inoculum was prepared from 48-hour cultures of C. flaccumfaciens var. aurantiacum grown on 2 percent potato-dextrose agar. The bacteria were removed from the agar surface and suspended in water and one aliquot equivalent to the bacterial growth on 1/4 Petri dish was applied to each pot at the time of transplanting as was the nematode inoculum. Fifty ml of bacterial suspension was added to the surface of the soil. Following inoculations the experimental material was kept in a greenhouse with air temperature maintained at about 22°C, which is favorable for development of bacterial wilt symptoms. The flats containing the different treatments were about 12 inches apart to prevent contamination. The plants were supported by stakes placed outside the pots to avoid possible damage to the roots.

Groups of 12 uniformly sized bean seedlings each received, unless otherwise stated, one of the following standard treatments at time of transplanting: 1) control, receiving neither pathogen; 2) bacterial inoculation; 3) root wounded; 4) root wounded plus bacterial inoculation; 5) nematode inoculation; 6) bacterial plus nematode inoculation. Root wounding of plants in treatments 3 and 4 was accomplished by pricking with a common pin while the roots were submerged in a bacterial suspension or in distilled water, depending upon the treatment. Tests were designed as randomized blocks; four replications of each treatment were employed.

Periodic readings were taken on the number of wilted or dead plants, and on height of plants. At harvest time, data included the total yield of seed and the number of seeds visibly infested by the orange-colored bacterial pathogen.

EXPERIMENTAL RESULTS

In the greenhouse experiment, as shown in Table 1, root wounding favored entrance of the wilt organism into the plant with 86 percent of population wilted in contrast to 6, 8, and 30 percent for the control, root wounded, and bacterial treatments, respectively. Because of the high incidence of bean wilt in the bacterial treatment without root wounding, the roots of the wilted plants were stained in hot lacto-phenol and a high population of root-lesion nematodes

Table 1. Percentage of wilted U.I. 59 bean plants with and without root wounding following addition of Corynebacterium flaccumfaciens var. aurantiacum in irrigation water in greenhouse tests in 1954.

Treatment	Distance from point of addition of bacteria[a]		
	1 - 8	9 - 16	17 - 24
Control	2	4	0
Root wounded	2	6	0
Bacteria	16	12	2
Root wounded plus bacteria	46	30	10

[a] In feet.

(Pratylenchus sp.) were discerned in them. The assumption was made that perhaps nematodes provided a portal of entry as a result of their feeding and penetration into the roots. Obviously wilting resulted from other causal agents as evidenced by a small percentage of wilting in uninoculated treatments. Care was taken not to contaminate these treatments with the bacterial organism during irrigation. Visible infection was not noted in the seed from any of the wilted plants. Isolation from stems of wilted plants revealed the presence of the orange-colored bacterium.

The pathogen can be disseminated by irrigation water with a decrease in percentage of infection as the distance from locus of infestation increases.

Attempts to substantiate these data under field conditions were not successful. Three such field tests were conducted for 3 years, 1954, 1955 and 1957. No evidence of dissemination or infection resulted either with or without wounding of roots. Also, 50 pods per row were selected at random and no visible seed infections were evident. These field tests were made with corresponding tests with the common blight organism, Xanthomonas phaseoli (E. F. Sm.) Dows., but negative results were obtained insofar as dissemination by irrigation water and root wounding are concerned.

Some exploratory tests were conducted in 1952 and 1955 to determine the effect that M. hapla Chitwood might have on the incidence of bacterial wilt of bean. The same strain of C. flaccumfaciens var. aurantiacum was used as was used in subsequent experiments. The nematode has been maintained on tobacco plants in the greenhouse. Observations were made on wilting and visible seed infection. Neither symptom appeared. Since only one egg sac per pot was used in the 1952 experiment, it was assumed that the amount of nematode inoculum was insufficient.

In 1955 the amount of inoculum of the root-knot nematode (M. hapla) was increased fivefold. Again no visible seed infection was evident. However, the yield of seed was decreased from 63 grams per replication for the control to 33 grams for the combination of nematode and bacteria. The average height of the plants at time of harvest for the control, bacteria, and nematode plus bacteria was 43, 39, and 27 inches, respectively. Since only a slight amount of galling was evident it was decided to employ another root-knot nematode species and two bacterial inoculation dates instead of one at planting time as used in the preliminary tests with M. hapla.

In the experiments reported in more detail in this paper, the nematode M. incognita was employed because it appears to be more virulent on beans, causing more severe galling.

In Table 2 are entered the data from Experiment 1 in which 10 egg sacs of M. incognita were applied per pot. Wilt occurred in the bacterial treatment (25 percent) but in lesser amounts than in the treatment in which the roots were wounded (37 percent) at time of bacterial inoculation or for the combination of nematodes plus bacteria (50 percent). Average yield was decreased slightly for the various treatments in which bacteria or nematodes were involved, with the combination of nematodes plus bacteria inflicting the most damage. Visible seed infection with bacteria resulted in only one treatment in which the roots were wounded at time of inoculation. Results from Experiment 2, shown in Table 3, demonstrate again that seed infection did not result based on macroscopic observation. Seed yield was decreased considerably when a combination of nematode plus bacteria was employed, resulting in a yield loss of 65 percent. Bacterial treatment caused a 50 percent loss in yield and 27 and 57 percent decrease for the nematode and bacteria plus root wounded treatments.

Table 2. Effect of bacterial wilt in U.I. 59 bean plants inoculated with Corynebacterium flaccumfaciens var. aurantiacum, with and without root wounding, and in combination with root-knot nematodes. Experiment 1.

Treatment	Percentage of plants wilted	Seed	
		Average yield[a] (grams)	Visibly infected with bacteria (number)
Control	0	76	0
Bacteria	25	63	0
Root wounded	0	74	0
Root wounded plus bacteria	37	68	12
Nematodes	6	62	0
Nematodes plus bacteria	50	57	0

[a] Average yield per replication.

Table 3. Effect of bacterial wilt in U.I. 59 bean plants inoculated with Corynebacterium flaccumfaciens var. aurantiacum, with and without root wounding, and in combination with root-knot nematodes. Experiment 2.

Treatment	Percentage of plants wilted	Seed	
		Average yield[a] (grams)	Visibly infected with bacteria (number)
Control	0	63	0
Bacteria	92	31	0
Root wounded	0	64	0
Root wounded plus bacteria	86	27	0
Nematodes	0	46	0
Nematodes plus bacteria	8	22	0

[a] Average yield per replication.

DISCUSSION

The experimental results presented in this paper do not follow a consistent pattern. In greenhouse experiments it was shown that the wilt organism can be carried in irrigation water for a distance of 24 feet, as evidenced by infection of bean plants. However, in three comparable field experiments conducted over a 3-year period wilting did not result following bacterial infestation of irrigation water. Wilt incidence was increased as a result of mechanical wounding of the roots, but plants with roots not so wounded also showed a fairly high amount of wilting. The roots from these plants were infected with root-lesion nematodes. The main objective of this paper was to test the hypothesis that nematodes may be instrumental in providing portals of entry for the wilt bacteria. However, root-knot nematodes were employed instead of root lesion nematodes because of an adequate supply of inoculum of the former pathogen.

The attempt to determine whether or not feeding of root by root-knot nematodes favored bacterial infection proved inconclusive. In one experiment the addition of nematodes actually decreased the percentage of plants showing wilt. In fact, in this experiment it would be difficult to increase the amount of wilt over the treatment in which bacteria were applied to the soil, since wilting occurred in 92 percent of the plants. Galling in the nematode treatments was very

severe, being present even on the stems. It must be stated that the amount of nematode inoculum -- 9 grams of galled tissue per pot -- might have been too great for a good test. An accurate count of the number of egg sacs was not made but it is assumed to be greater than 10 egg sacs, based on severity of galling on roots and stems.

In the second experiment 10 egg sacs per pot were used and may have given a more accurate picture. The number of wilted plants was increased by root wounding plus bacteria and nematode plus bacteria treatments. The former treatment increased the number of wilted plants by 50 percent and the latter treatment doubled the number over the control. In both experiments, wilting was not a symptom of nematodes alone, perhaps because the air temperature was not as high as out-of-doors.

When a high amount of nematode inoculum was used yield was decreased by 27 percent, compared with the checks. A similar trend was noted when less nematode inoculum was used.

In all the experiments it is quite evident that seed infection as detected macroscopically is not an adequate criterion in judging the wilting rates induced by different types of treatments. In fact, in only one instance was a small percentage of seed visibly infected. This was the bacteria plus root wounding treatment.

The combination of nematode plus bacteria unually caused a necrosis of the roots. The bacterial organism considered as a vascular parasite may be able to induce necrosis under these circumstances. This does not preclude the possibility that an air-borne organism may have entered the soil over a 3-month period and been instrumental in inducing browning of the roots.

In the experiment in which bacteria apparently were prevented from entering the vascular system, thus causing wilting, there may have been a morphological or physiological effect. The nematodes induce rapid cell division and proliferation and may prevent the bacteria from entering the vascular tissue, thereby hindering the establishment of infection. Physiological changes may have been initiated in the bean plants as a result of heavy nematode infection of the root and stem areas. Nematodes are capable of effecting a change in the host as evidenced by chlorosis, stunting, and other symptoms.

The fact that bacterial wilt occurred without mechanical wounding of the roots does not rule out the possibility that the organism is a wound parasite. Perhaps the natural wounding resulting from development of secondary roots induces sufficient injury for the organism to penetrate. Possibly the mere penetration of the soil by the roots may inflict self-injury sufficient under the circumstances to favor penetration and development of the wilt bacteria into the root. The author is unable to explain why in an earlier report (3) wilt bacteria in sterilized soil in which beans were grown did not cause wilting, but did cause wilting in comparable tests in nonsterilized soil.

Perhaps under certain inoculum potential levels of root-knot nematodes the evidence in this study would indicate merely a wounding effect on the part of the nematodes which is in agreement with the findings of Stewart and Schindler (4) and of Lucas, Sasser, and Kelman (2). Kelman (1) reports the Granville wilt bacterium as a wound parasite of tobacco, susceptible varieties being invaded through natural wounds in the roots. Stewart and Schindler (4) also suggest that infection by bacteria could occur through a small number of natural breaks in the root system.

There was no apparent direct effect of mechanical root wounding of beans in these tests, as evidenced by the yield data. Perhaps differences due to variation in pathogenicity of the two nematode species may have a decided effect on the rate of wilting in the presence of bacteria. Stewart and Schindler (4) report such differences in nematodes they employed, with M. hapla inducing a lower rate of wilting than M. incognita. The present study showed a similar result with these two species. Perhaps it was due to a greater concentration of inoculum of M. incognita or to a greater pathogenicity for the host variety employed. M. hapla produced small galls whereas M. incognita induced many large galls on the roots of beans.

The role that nematodes may have in inducing bacterial wilt of beans under field conditions is problematical. Since both pathogens are present in the bean-growing area of the State, such a relation may exist. Although the wilt disease of beans is more widespread in a field under hailed conditions, circumstantial evidence indicates that other factors such as cultivation damage, nematodes and natural wounding may cause sufficient injury to the roots for entrance of the bacteria into the plant.

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RELATIONS BETWEEN NEMATODES, FUMIGATION AND FERTILIZATION IN RICE CULTURE

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Abstract

Soil fumigants (Dowfume MC-2, D-D) and fertilizers (P, P + K, N + P + K) exerted independent, additive effects on rice yields, with one exception. Phosphorus reduced growth in plots fumigated with Dowfume MC-2. This was believed caused by a retardation of reducing activities following submergence. The addition of ammonium nitrogen eliminated the phosphorus effect. Fumigants stimulated plant growth by hygienic and nutritive effects, only the latter could be partially replaced by fertilizers. The influence of nematodes was measured within the hygienic effects of soil fumigation. Results of fumigation, greenhouse pathogenicity tests and feeding observations of Tylenchorhynchus martini on rice roots indicated that hygienic effects of soil fumigation did not result from elimination of T. martini alone, or a single complex of nematodes and other microorganisms, but from suppression of unknown injurious and competitive soil factors.

Preliminary soil fumigation experiments (2, 4, 6) demonstrated that certain plant parasitic nematodes infest rice and that one of four tests yielded a crop response correlated with reductions of nematode populations. It is axiomatic that such variability could result from fumigant-nutrient element interactions and their intensification by the semi-anaerobic soil environment during crop development (10, 11, 12).

There are at least two growth stimulating effects in soil fumigation tests: a) effects of fertilizers and interactions with fumigants, b) hygienic effects. The first effect is complex and may be induced directly by the fumigant. Goffart and Heiling (5) found a specific effect of chlorine ions on sugar beets in plots treated with D-D. This was a nutritive effect, manifested as a more rapid recovery from wilting and correlated with factors related to water and ash content of tissues. Aldrich and Martin (1) showed that partial sterilization of soils by fumigation altered the proportion of ammonium to nitrate nitrogen but not the total amount of the two forms. The authors found that crop stimulation (excluding hygienic effects) was not due to increased nutrient availability in the soils used. They reasoned, however, that such effects might be produced in other soils. Thus, the nutrient supply might be raised from a deficient to sufficient level in some soils, and in others, from an adequate to a toxic level, resulting in decreased plant growth. Fumigation increased the quantities of soluble cations and anions and they concluded that the observed stimulation in plant growth was caused by direct chemical reactions between fumigant and soil.

Any indirect nutritive effect of soil fumigation resulting from dead microorganisms is generally excluded by the well-known, partial effect of soil treatment. It has long been known that the near destruction of microorganisms by fumigation is followed by large increases in numbers upon incubation.

The hygienic effect of soil fumigation is considered most important because its primary purpose is to eliminate nematodes and other injurious and competitive microorganisms. The idea that soil fumigant effects can be analyzed in terms of plant responses to nutritive factors is meritorious, as a partial solution of the problem, provided the hygienic aspects of soil fumigation are given proper weight. Plant responses unassigned, after a complete assessment of the fertility and nutritive effects of soil fumigation, must be due to the hygienic benefits resulting from elimination of nematodes and other competitive and injurious factors.

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Nematologists have provided plant pathologists with a new concept, applicable to pathogen-suscept relations; namely, the idea of nematodes as catalysts or activators of plant diseases. This hypothesis should probably be stated in the simple terms of nematodes as agents of mechanical injury which provide infection courts for more potent microorganisms. The evidence restricts association of nematodes and other microorganisms in identifiable disease complexes to those forms which, singly, are capable of pathogenic effects on higher plants. That disease might ensue from associations of nonpathogenic, parasitic or other types of innocuous microorganisms is a stimulating idea, if confined to an individual complex; it becomes depressing in terms of many complexes or a multiplicity of factors. Depressing or not, this is the only hypothesis involving biotic factors which has not been negated at least once by the results of nematode or soil fumigation experiments in rice. Unfortunately it cannot be subjected to critical tests and gains only when another hypothesis is unsupported.

The purpose of this paper is to discuss nematode-rice relations in Louisiana and Texas on the basis of experimental data and observations from one field experiment, numerous greenhouse experiments, and the photomicrography of Tylenchorhynchus martini Fielding 1956 feeding on rice roots in agar.

SOIL FUMIGANT-FERTILIZER EXPERIMENT

This experiment was conducted in 1956 on the Louisiana State University Rice Experiment Station at Crowley in cooperation with personnel of that station. The soil was a Crowley Silt Loam (6, 11) of low fertility, on which rice commonly makes a strong response to nitrogen and weak responses to phosphorus and potassium fertilizers. Recommendations (12) in pounds per acre of fertilizer constituents for silt loam soils are: N, 40 to 60; P_2O_5, 20 to 30; K_2O, 0 to 30.

Fumigant plots, 20 feet long and 34 feet wide, were in a 3 x 3 Latin square and separated rowwise by 10-foot buffers and columnwise by 6-foot buffers. Fumigant plots were subdivided into four fertilizer plots, each 7 feet wide and 20 feet long, and separated by 2-foot buffers.

Temperature, moisture and mechanical condition of the soil were favorable for fumigation on April 6 with halogenated hydrocarbon nematocides[2]. D-D was applied at 42 gallons per acre with a MacLean hand applicator at a 6-inch depth on a grid pattern with injection points at intervals of 1 foot. Dowfume MC-2 was applied under plastic covers at 3 pounds per 100 square feet from 1 pound cans fitted with adapters and plastic tubing. The plastic covers were retained on the plots 48 hours.

Fertilizers, broadcast with an applicator at planting time, were ammonium sulfate, superphosphate, and muriate of potash. Fertilizer treatments in pounds per acre of N, P_2O_5 and K_2O, respectively, were: a) 0:0:0, b) 0:40:0, c) 0:40:40, d) 60:40:40. Thus, there were 3 fumigant treatments - no fumigation, D-D and Dowfume MC-2, and 4 fertilizer treatments = 12 combinations. Each was replicated 3 times, for a total of 36 plots in the experiment.

Toro, a long grain rice, was planted April 17. The site was flooded with water from May 1 to August 15; except for a 10-day period during the first two weeks in July, when rainfall maintained soil saturation. Each plot was harvested September 1 by cutting plants 2 inches above the soil line in 7 x 7 foot squares and weighing the shocks for total forage plus grain. Responses to fertilization and soil fumigation were analyzed in terms of total forage, grains yield and quality. Yield values were converted to pounds per acre (Table 1).

Total free-living and plant parasitic nematodes were determined from 1 pint soil samples, taken from each of the nine fumigant treatment plots at five different intervals during the growing season. The samples were processed, nematodes enumerated and data evaluated in terms of sampling fluctuations (6, p. 17). T. martini and Radopholus oryzae (V. Breda de Haan 1902) Thorne 1949 occurred in extremely small numbers and only R. oryzae showed significant population fluctuations between samplings. The complete data are in Table 2, where numbers per 1/7 pint of soil are averaged for three replicate plots in each treatment. Low numbers of nematodes in submerged soil of the rice test resulted from population reductions by microbiological factors (6, 7, 8, 9). An adjacent area of white clover yielded 20 times as many T. martini in comparable soil samples.

Yields of rice are partitioned in Table 3, according to causal effects. Thus, the yields of total forage and grain due to fumigant, fertilizer and combined effects are calculated as per-

[2] These previously described materials (6) were supplied by the Shell Chemical Corporation of New Orleans, Louisiana and the Dow Chemical Company of Greenville, Mississippi.

Table 1. Effects of soil fumigation and fertilization on Toro rice yields at Crowley, Louisiana in 1956.

Soil treatment)	Fumigant	Fertilizer[a] Pounds per acre of N, P, K)	Pounds per acre Forage plus grain	Grain
1	Check (No	0- 0- 0	3200	1580
2	treatment)	0-40- 0	4089	1826
3	"	0-40-40	5245	2199
4	"	60-40-40	10935	3615
5	D-D (42 gal./A.)	0- 0- 0	5779	2213
6	"	0-40- 0	5512	2299
7	"	0-40-40	6934	2772
8	"	60-40-40	14402	4450
9	Dowfume MC-2	0- 0- 0	12979	4246
10	(3 lbs/100	0-40- 0	11113	4018
11	sq. ft.)	0-40-40	12090	4090
12	"	60-40-40	19825	5176

[a] N was calculated as N, P as P_2O_5 and K as K_2O.

Table 2. Plant parasitic nematodes in 1956 rice experiments at Crowley, Louisiana (numbers per 1/7 pint of soil, averages of 3 replicate plots).

Fumigant	Sampling 3 May	6 June	13 July	8 Aug.	7 Sept.
			Tylenchorhynchus martini		
Check	7	4	1	2	< 1[a]
D-D	0	3	0	< 1	3
Dowfume MC-2	0	1	0	0	< 1
			Radopholus oryzae		
Check	1	10	5	< 1	< 1
D-D	0	5	1	< 1	< 1
Dowfume MC-2	0	< 2	2	< 1	< 1

[a] < means less than and obviates the need for decimals.

centage increases over designated checks (boxed values). The effects of fertilizers on rice in nonfumigated soil was demonstrated by low responses to phosphorus and phosphorus with potassium (treatments 2 and 3), and a marked response to nitrogen (treatment 4). Parallel trends developed in plots fumigated with D-D (treatments 5-8) and with Dowfume MC-2 (treatments 9-12), except that yields were depressed by phosphorus in phosphorus and phosphorus and potassium treatments. Fertilizer levels in the experiment exceeded recommendations for average rates of application and fertilizer substituted for much of the fumigant effect

Table 3. Percentage increases of Toro rice resulting from soil fumigation and fertilization at Crowley, Louisiana in 1956.

Soil treatment	Percentage increases[a] due to: Fumigants — Forage plus grain	Fumigants — Grain	Fertilizers — Forage plus grain	Fertilizers — Grain	Total — Forage plus grain	Total — Grain
1	/3200/	/1580/	/3200/	/1580/	/3200/	/1580/
2	/4089/	/1826/	28	16		
3	/5245/	/2199/	64	39		
4	/10935/	/3615/	242	129		
5	81	40	/5779/	/2213/		
6	35	26	-5	4	72	46
7	32	26	20	25	117	75
8	32	23	149	101	350	282
9	306	169	/12979/	/4246/		
10	172	120	-14	-5	247	154
11	130	86	-7	-4	278	159
12	81	43	53	22	520	228

[a] Percentage increases were calculated in terms of checks (boxed values which are in pounds per acre). Fumigant effects were determined at equal fertilizer levels; for example the percentage increase for treatment 7 (32) was calculated in terms of treatment 3 (5245).

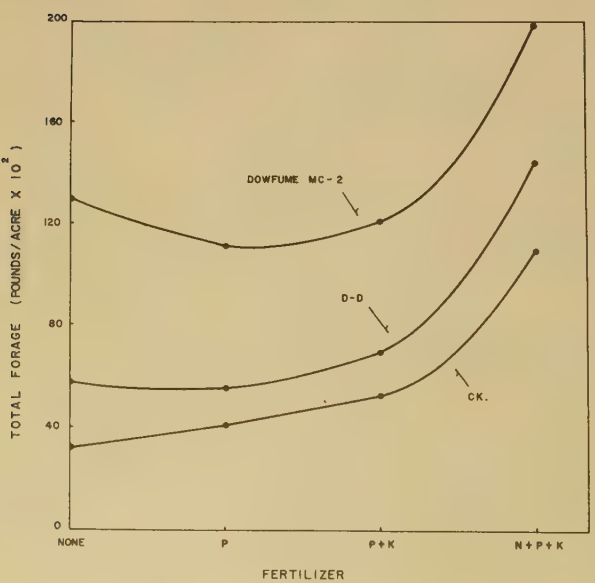

FIGURE 1. Responses of Toro rice to soil fumigants (D-D and Dowfume MC-2) at Crowley, Louisiana in 1956. (Fertilizer treatments in pounds per acre, respectively of N, P_2O_5 and K_2O were: a) 0:0:0, b) 0:40:0, c) 0:40:40, d) 60:40:40)

(treatments 8, 12). Percentage increases in yields due to fumigation diminished progressively with fertilizer increment. Phosphorus alone appeared to diminish the effects of D-D, but all fertilizers diminished the effects of Dowfume MC-2.

Yield responses to fertilizers in the different fumigant treatments are shown in Figure 1. Common shape of curves indicates independent effects of fumigants and fertilizers on yields. However, there was a depression of yields by phosphorus, related to fumigant effect. Percentagewise, fumigant effects on yields were greatest at no fertilizer and diminished up the scale of fertilizer additions (Table 3). However, actual yield increases due to fumigants, at different fertilizer levels, were generally equivalent (Figure 1).

Milling and iodine test results (Table 4) indicated no relation between fumigant treatment and grain quality.

Table 4. Quality tests[a] on rough Toro rice in the experiment at Crowley, Louisiana in 1956.

Soil treatment	Milling test: Percentage whole grain	Milling test: Percentage whole plus broken grain	Iodine test for cooking quality
1	66.23	70.36	62.8
2	66.00	70.73	62.0
3	66.13	70.03	65.7
4	65.13	70.02	70.0
5	63.03	70.43	56.3
6	66.33	70.53	59.3
7	66.16	70.23	58.5
8	66.63	71.53	66.2
9	64.80	69.86	69.0
10	65.43	69.90	68.2
11	64.73	70.00	62.0
12	63.93	71.00	66.7

[a] The tests were conducted by Dr. John V. Halick, Chemist, A. R. S. Values are averages of 3 replications based on cleaned samples at approximately 13 percent moisture.

FEEDING OF T. MARTINI ON RICE ROOTS

Feeding of T. martini on rice roots has been observed and photographed by the senior author. Chambers were constructed of laboratory glassware. A glass ring was cemented to a large glass slide and covered by a cylinder plugged with cotton. Rice seedlings, produced by germination of seeds in Petri plates, were imbedded in 3 percent agar in water in the ring, then inoculated with heavy suspensions of T. martini and covered with the cylinder to preserve humidity. Observations and photographs were made after removal of the cylinder and inversion of the chamber on the microscope stage.

Under the imposed cultural conditions, population increase of T. martini was rapid, and in cultures maintained for periods up to 3 weeks numerous eggs and new individuals were observed. Nematodes fed at random positions on the root, and at variable depths of penetration, ranging from the epidermis to the stele. A photograph (6) shows two individuals in the meristematic region of a root, and a nematode is in the root hair region (Figure 2). Feeding at root tips was common but root tips were not preferentially selected. Feeding was most prevalent on growing roots but was observed on roots apparently hypostatized by lack of nutrients. Penetration of roots, by destruction of cells and portions of cells, was in a straight line in the path of the nematode. Numerous photographs (not shown) demonstrated clean tunneling without collapse of adjacent cell walls. Funnel diameters approximated nematode body diameters. There was no discoloration, necrosis or pectolysis of root tissues as a result of penetration and feeding by T. martini.

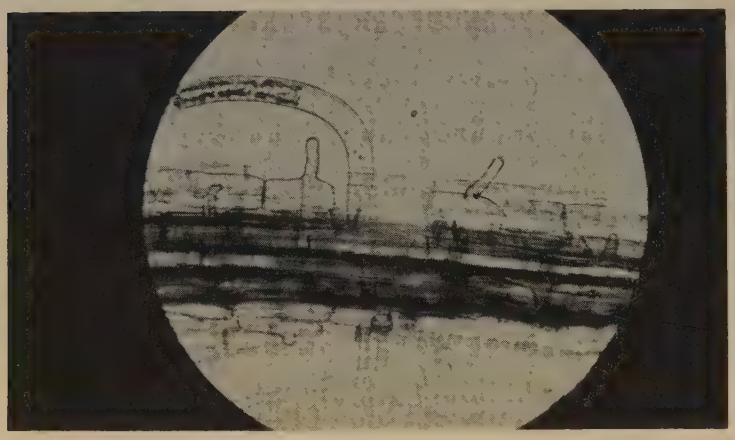

FIGURE 2. Tylenchorhynchus martini Fielding 1956 in feeding position with head penetrating the stelar position of the root hair zone of a rice root.

PATHOGENICITY TRIALS WITH T. MARTINI IN STERILIZED SOIL

Several greenhouse experiments were conducted in moist soils, comprising Crowley Silt Loam and mixtures of Mississippi terrace and river bottom soils and sand. T. martini, in picked or relatively-pure mixed populations, was added in large numbers to pots of steam-sterilized soil planted to rice. Cultures, including checks without nematodes, were watered to maintain average moisture levels. Populations of T. martini in the tests rose frequently to more than 2000 per pint of soil within 6 weeks but growth of rice was not significantly affected.

The design of greenhouse tests was changed after the 1956 field experiment to provide for submergence of cultures in water. A phycomycetous fungus in water-saturated soil might have caused, in association with T. martini, some of the observed growth and yield reductions in the field experiment. Nematodes and other microorganisms were added to sterilized Crowley Silt Loam in 10-quart plum boxes, which were submerged in water in galvanized steel tanks to simulate field conditions. Six experiments were conducted with heavy plantings of rice (20 to 25 per box) in some instances and, in others, light plantings (2 to 3 plants per box, transplanted from Petri plates). Varieties of rice were Zenith, Nato and Toro. A typical design was: a) nonsterile check, b) sterile check, c) nemas (2500 T. martini per box), d) Pythium spinosum[3] Sawada, and e) nemas plus fungus. The soil was sterilized in tank tests with Dowfume MC-2. In two tests the fungus was added in rice roots from nonsterile field soil. (Lindberg and Hollis observed and stained phycomycetous hyphae in cortical tissues of rice roots from nonsterile field soil.)

There were slight, early effects on growth of rice plants in these experiments attributable to nematodes and fungi. However their magnitude and persistence were not sufficient to merit comparison with the 1956 field results.

DISCUSSION

Initial soil fumigation experiments in Louisiana (6) were designed on the premise that maximal yield responses to fumigation should result at minimal fertilizer levels. Maximal fertilizer, on the other hand, should "repair" or compensate for the effects of nematodes in check plots and, at the same time, should "mask" fertilizer effects of soil fumigation, thus reducing treatment differences. The minimal fertilizer approach, tested in 12 experiments in 1955, increased the percentage differences between treatments and emphasized the effects of soil fumigation but it contributed no basic information on responses of crops to fumigants. The present results show that absolute differences in crop response resulting from fumigant treatments are fairly constant, regardless of fertilizer treatment (Fig. 1).

3 Dr. G. D. Lindberg isolated P. spinosum from rice seedlings growing in field soil and has cooperated in all tank experiments.

The alternative hypothesis, that maximal fertilizer levels should result in maximal differences in fumigant effects, has contributed knowledge of soil fumigation and crop responses. It may be argued that a plant growing in fumigated soil should utilize higher concentrations of plant nutrients through a more efficiently absorbing root system, enhancing the effects of soil fumigation. Consequently, the nutrient element content of plants, growing in fumigated as compared with nonfumigated soil, should be higher per unit weight of plant tissue. This possibility was tested in soil fumigation experiments at the Louisiana State University Branch Experiment Station at Homer in 1955-56 (18). Tissue analyses for both major and minor nutrient elements in several crops demonstrated that nutrient levels in vegetative plant tissues were not raised by soil fumigation. There was, therefore, no synergism between soil fumigation and fertilization. A plant in fumigated soil possesses a more extensive root system and utilizes more nutrients, by virtue of its greater mass, but does not differ in the quality of utilization per unit of root tissue. Therefore, soil fumigation and fertilization are additive in their effects on plant growth.

Thorne (13, 14) has pointed out the disturbing effects of plant parasitic nematodes on agronomic and horticultural investigations. If nematodes were uniformly distributed around crop plants and fumigant and fertilizer effects did not interact, nematode disturbance would be uniform and deductible from field trials. Plant parasitic nematodes appear uniformly distributed in rice fields (6, p. 8), and present data are substantially free of fumigant-fertilizer interactions. However, the practice of deducting nematode effects would be premature, even in rice, because of lack of analytical data and the complexity of the soil as a locus of physical, chemical and biological activity.

The results of this work suggest that fertilizer treatments cannot make up completely for fumigant effects. That portion of the plant growth stimulation which may be substituted by fertilizers is interpreted subjectively as a fertilizer effect of soil fumigation; the remainder is considered a hygienic effect. Soil and plant analyses could supply data for a more precise measure of this "fertilizer equivalent" of soil fumigation.

A coherent pattern of rice fertilizer requirements has been derived by Ponnamperuma (10) in his study of the chemistry of submerged soils. In practice, ammonium nitrogen is generally superior to nitrate nitrogen for rice. This stems from a variety of conditions, including loss of nitrate by leaching and denitrification, and retardation of reduction processes in acid soils by nitrate. This latter factor is not operative in Crowley Silt Loam, which is near neutral or alkaline in reaction (11). The iron requirement of rice is higher than for most crop plants and the reduction of iron and manganese is almost entirely a result of the anaerobic metabolism of soil bacteria. According to Ponnamperuma, this is the outstanding beneficial effect of submergence. Since nitrate fertilizer was not added in the Crowley experiment, it is probable that sufficient nitrate was not present to retard reduction processes. Nevertheless, fumigation may have slowed reducing processes by its effects on populations of reducing bacteria. Sturgis (11) found that solubility of phosphorus was markedly lowered as reducing conditions developed in Crowley soils. The inhibitory effect of phosphorus on rice in plots fumigated with Dowfume MC-2 suggests that this chemical treatment retarded reduction processes and growth at insufficient nitrogen. Addition of ammonium nitrogen to the fertilizer overcame this growth inhibition.

Yield responses of rice to Dowfume MC-2 and D-D showed the former to be two to three times more effective. Such results are a basis for the working hypothesis that a greater yield response to Dowfume MC-2 over D-D indicates that a complex of nematodes plus another microorganism reduces yields in nonfumigated soils. This hypothesis is a good starting point for greenhouse tests but not for explaining the results of field experiments (2).

T. martini produced abnormally blunt, irregular and sparse roots in sugarcane plants developing from seedpieces, when added in large numbers to sterilized soil in clay pots (3); however significant reductions in top growth of sugarcane did not occur. Field, greenhouse and laboratory data indicate that T. martini is a nonpathogenic parasite on rice.

The position of R. oryzae as a pathogen on rice is ambiguous. Van der Vecht (15) and Van der Vecht and Bergman (16) concluded that R. oryzae is parasitic and injurious to rice in Java, under growth conditions which reduce the capacity of rice plants for recovery. Their data suggest that R. oryzae is not intrinsically pathogenic to rice, and that damage may result only from attacks by exceedingly large numbers of the nematode. Whitlock (17) concluded that R. oryzae probably does little damage to rice in Louisiana and Texas. It appears that R. oryzae may damage rice in Java under cultural conditions which are not operative in Louisiana and Texas. A multiplicity of injurious and competitive factors must be invoked at present to explain the hygienic effects of soil fumigation on rice in Louisiana and Texas.

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SAMPLING "PULLED AND TREATED" AREAS FOR THE BURROWING NEMA RADOPHOLUS SIMILIS (COBB) THORNE

Wray Birchfield, W. G. Cowperthwaite, C. Poucher, and J. M. McNamee[1]

INTRODUCTION

Spreading decline of citrus (5, 14) caused by the burrowing nema (used throughout manuscript according to Chitwood (4), Radopholus similis, for several years has been considered the most important citrus disease in Florida. The only known control measure is the so-called "pull and treat" method recommended by Suit et al. (15, 16). By use of this method all infected trees plus a margin of healthy trees, for safety, are removed from a diseased area and burned; the remaining roots are raked and destroyed as well as possible. The soil is then prepared and fumigated with DD at 60 gallons per acre injected to a depth of 12 inches. The first two rows of trees beyond the known infestation are removed as a safety factor in areas where the entire grove is not diseased or where marginal areas of infestation are involved. A waiting period of 2 years before replanting was suggested so that the margins could be checked for infected trees that might have been left at the time of delimitation. Subsequent work (1) indicated that the burrowing nema could survive only about 4 months without food, further justifying a waiting period and encouraging clean cultivation of the "pulled and treated" areas.

PULLED AND TREATED PROPERTIES

In September of 1955 the State Plant Board of Florida launched a program to use the "pull and treat method" as developed by the Citrus Experiment Station and advocated by the industry. The impossibility of delimiting the margins of infestation on the basis of symptoms and the low populations of the burrowing nema often encountered on the margins rendered the work difficult. Other impediments encountered in the program were court litigations, freezes, and awaiting the removal of crops before destroying the trees. Nevertheless, by October 1, 1958, 573 properties representing about 3736 acres had been removed, as shown in Table 1. This was about half of the estimated decline acreage in the State.

Table 1. Shows the spreading decline properties "pulled and treated" and the acreage involved.

Counties	Number of properties	Acreage
Charlotte	1	40.00
DeSoto	1	7.00
Highlands	104	117.75
Hillsborough	5	115.75
Lake	47	252.00
Orange	23	134.25
Pasco	1	10.00
Pinellas	2	10.00
Polk	389	3049.67
Totals	573	3736.42

MARGINAL INSPECTIONS

In 1957 a survey was made of "pulled and treated" areas having margins to determine whether or not delimiting of the infestations was successful. At first a single sample was taken from each marginal tree, but later three samples were taken from each tree in the first two rows. Two examinations of the margins were made and a third is in progress. About 554 properties received marginal inspections. The first inspection showed 96 positive out 313 examined, the second 58 out of 202, and the third 1 out of 39. This was a total of 155 positives

[1] Phytonematologist, Plant Commissioner, Assistant Chief Plant Inspector, and Special Inspector, respectively, State Plant Board of Florida.

out of 554 examined. Considered from the standpoint of total inspections, the margins were found about 28 percent infested. However, only about 3 percent of the trees along the margins were actually infected.

NEMA INSPECTIONS

In a preliminary survey, 2516 root samples from plants growing in 126 treated properties showed two samples of beggar weed (Meibomia sp.) positive to the burrowing nema. A second and more intensive survey of about 23 properties was made using both roots and soil from the pulled and treated areas. Besides direct root examinations, soil and roots were collected and processed by use of corn as a bioassay for the burrowing nema. Previous tests had shown that corn would recover the organism from soil when present in small numbers and that apparently it was one of the best plants to employ in the greenhouse for this purpose.

About 23 properties which had received treatment 2 years previously were reconnoitered for host plants from which to examine soil. The most commonly encountered were: Beggar weed (Meibomia spp.), Bermuda grass (Cynodon dactylon), Caesar's weed (Urena lobata), citron (Citrullus vulgaris citroides), crabgrass (Digitaria spp.), sandspur (Cenchrus spp.), and nutgrass (Cyperus rotundus). Several additional host plants were found less commonly, such as hairy indigo and nightshade (Solanum gracile). Instead of sampling the areas uniformly, spots were chosen where host plants were concentrated. When only the roots of plants were employed these were generally sampled as 3- to 4-foot depths.

Soil and roots were taken from 1-, 3-, and 6-foot depths in the immediate vicinity of a given host plant. Four gallons of soil were usually taken from each level, placed in the greenhouse and planted with sweet corn (Zea mays var. golden bantam x). After approximately 40 to 60 days the corn roots were processed according to the Young technique (17). A record of the plants examined and the nemas recovered is shown in Table 2.

Table 2. Shows kinds of plants examined from "pulled and treated areas" and nemas recovered.

Plants examined	Number of samples	Nemas	Percent positive
Beggar weed (Meibomia spp.)	27	Aphelenchoides spp.	7
		Aphelenchus avenae	11
		Criconemoides sp.	15
		Hoplolaimus tylenchiformis	4
		Meloidogyne spp.	22
		Pratylenchus brachyurus	26
		Pratylenchus zea	4
		Pratylenchus spp.	11
Bermuda grass (Cynodon dactylon Pers.)	428	Aphelenchoides spp.	19
		Aphelenchus avenae	7
		Aphelenchus spp.	2
		Belonolaimus sp.	-1
		Criconemoides sp.	8
		Dorylaimus spp.	-1
		Hoplolaimus tylenchiformis	20
		Meloidogyne spp.	1
		Pratylenchus brachyurus	18
		Pratylenchus zea	14
		Pratylenchus spp.	16
		Radopholus similis	-1
Caesar's weed (Urena lobata L.)	104	Aphelenchoides spp.	7
		Aphelenchus avenae	8
		Aphelenchus spp.	5
		Ditylenchus sp.	3
		Dorylaimus spp.	2
		Hoplolaimus tylenchiformis	6
		Meloidogyne spp.	6

Table 2 continued.

Plants examined	Number of samples	Nemas	Percent positive
		Pratylenchus brachyurus	3
		Pratylenchus zea	13
		Pratylenchus spp.	12
		Paraphelenchus sp.	4
		Tylenchus spp.	3
Citron (Citrullus vulgaris var. citroides, Bailey)	24	Aphelenchoides spp.	25
		Aphelenchus avenae	33
		Aphelenchus spp.	8
		Dorylaimus spp.	17
		Meloidogyne spp.	8
		Pratylenchus zea	21
		Pratylenchus spp.	8
		Tylenchus spp.	8
Citrus spp. (margin of areas)	419	Aphelenchoides spp.	2
		Aphelenchus avenae	6
		Aphelenchus spp.	3
		Belonolaimus sp.	5
		Criconemoides sp.	3
		Dorylaimus sp.	-1
		Hoplolaimus tylenchiformis	7
		Helicotylenchus sp.	-1
		Meloidogyne spp.	1
		Pratylenchus brachyurus	7
		Pratylenchus zea	-1
		Pratylenchus spp.	2
		Paraphelenchus sp.	-1
		Tylenchus spp.	2
		Tylenchus semipenetrans	3
Cottonweed (Froelichia floridana (Nutt.) Moq.	1	Pratylenchus brachyurus	100
Crabgrass (Digitaria spp.)	61	Aphelenchus avenae	5
		Criconemoides sp.	2
		Meloidogyne sp.	7
		Pratylenchus brachyurus	38
		Pratylenchus scribneri	10
		Pratylenchus zea	5
		Pratylenchus spp.	10
		Tylenchus sp.	2
Hairy indigo (Indigofera hirsuta L.)	15	Aphelenchus avenae	7
		Hoplolaimus tylenchiformis	7
		Pratylenchus brachyurus	93
		Pratylenchus sp.	7
		Radopholus similis	7
Lamb's quarters (Chenopodium album L.)	3	Aphelenchus sp.	33
		Nothotylenchus sp.	7
Lemon sprouts (Citrus limon Burm.)	489	Aphelenchoides spp.	4
		Aphelenchus avenae	8
		Aphelenchus spp.	5
		Criconemoides sp.	5
		Ditylenchus sp.	-1

Table 2 continued.

Plants examined	Number of samples	Nemas	Percent positive
		Dorylaimus sp.	-1
		Hoplolaimus tylenchiformis	-1
		Meloidogyne spp.	24
		Pratylenchus brachyurus	22
		Pratylenchus spp.	5
		Radopholus similis	3
		Tylenchus spp.	1
Mulberry (Morus rubra L.)	1	Meloidogyne sp.	100
		Pratylenchus zea	100
Nutgrass (Cyperus rotundus L.)	62	Aphelenchoides sp. 1	2
		Criconemoides sp.	12
		Meloidogyne sp.	2
		Pratylenchus brachyurus	10
		Pratylenchus zea	8
		Pratylenchus sp.	8
		Tylenchus sp.	13
Periwinkle (Vinca rosea L.)	10	Saprozoics only	
Sandspur (Cenchrus echinata L.)	24	Aphelenchus spp.	4
		Criconemoides sp.	38
		Pratylenchus brachyurus	33
		Pratylenchus zea	46
Silk oak (Grevillea robusta Cunn.)	5	Aphelenchus spp.	20
		Meloidogyne spp.	60
Teaweed (Sida carpinifolia L.)	15	Aphelenchoides spp.	13
		Aphelenchus avenae	13
		Criconemoides sp.	20
		Pratylenchus brachyurus	7
		Pratylenchus zea	7
		Pratylenchus spp.	40
		Tylenchus sp.	7
Unknown	172	Aphelenchoides spp.	19
		Aphelenchus avenae	17
		Aphelenchus spp.	11
		Belonolaimus sp.	-1
		Criconemoides sp.	4
		Dorylaimus spp.	2
		Hoplolaimus tylenchiformis	6
		Meloidogyne spp.	-1
		Nothotylenchus sp.	-1
		Paraphelenchus sp.	1
		Pratylenchus brachyurus	2
		Pratylenchus zea	1
		Pratylenchus spp.	9
		Trichodorus sp.	-1
		Tylenchinae (unknown)	-1

Radopholus similis was found in four out of 23 fumigated properties examined. In all cases it was recovered by the corn technique. It was found by direct root examination in one property. Considered from the standpoint of properties examined, about 17 percent were positive;

placed on a sample basis, this figure was slightly less than 1 percent positive.

The plant-parasitic types recovered approximately 2 years after fumigation with DD at 60 gallons per acre were those of the natural citrus soils (6) in Polk County for the most part. Radopholus similis was found in relatively few properties, compared with Pratylenchus spp. and Meloidogyne spp., which were found in most of the properties examined. This suggests that R. similis is perhaps not as competitive and adaptive to a rapidly changed environment, including feeding conditions, as are the others. However, there are undoubtedly several factors involved, so that a comparison of any two given nemic species cannot be made without full qualifications. It is also conceivable that R. similis is more susceptible to DD than are other plant-parasitic forms, but specificity of fumigants is generally not considered a key factor. Nemas associated with specific plants are listed in Table 2. It should be pointed out that the technique involved was directed primarily for the burrowing nema, which is an endoparasite; consequently, the number of ectoparasitic forms recovered by this technique was surprising. Pratylenchus brachyurus and P. zea were very common in the pulled and treated areas. P. brachyurus is believed to be a damaging parasite of citrus but thorough studies have not been made; however, P. zea is believed to feed mostly on grasses and herbaceous plants in the groves.

CITRUS SPROUT SURVEY

Lemon sprouts were found in about half the properties surveyed for the burrowing nema. It was conceivable that the burrowing nema existed in all these plants and, therefore, a further survey was made for citrus sprouts in 404 additional properties. Sprouts were found in 70 out of the 404 properties examined. These sprouts were destroyed and the immediate area refumigated.

Failure to destroy all the citrus sprouts in a given area was believed to be caused by mechanical failure of the fumigating equipment, such as running out of DD or clogging of spray nozzles since this material is phytotoxic to citrus. In all properties where lemon sprouts were found, clean cultivation had not been practiced to a significant degree. Few or no citrus sprouts were found in those areas where clean cultivation had been practiced. Re-emphasis should be made of the importance of keeping the "pulled and treated" areas fallow for at least 4 to 6 months.

DISCUSSION

The data collected in regard to the burrowing nematode and citrus sprouts found in "pulled and treated" areas should serve as a basis for the evaluation of the "pull and treat" program. In areas with successfully delimited margins, where clean fallow had been practiced at least 6 months and no citrus sprouts left in the area, the burrowing nema was not recovered. It would seem, from the fact that burrowing nemas were recovered in some properties, that a few generally escape fumigation, either by failure of the chemical to penetrate deeply enough or by failure of fumigating apparatus. Nevertheless, other factors, such as parasitism, starvation, or failure of the burrowing nema to adapt quickly to a new environment, are evidently in favor of the program. The "pull and treat" program would appear to be basically sound in regard to containing and slowing the progress of the disease. The large percentage of "pulled and treated" areas in which the burrowing nema could not be detected indicated that this operation has considerable merit. However, a tighter regulation regarding complete citrus root removal and fallow would greatly enhance the program. A complete evaluation must, of necessity, await the sampling of citrus replanted in these areas.

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STATE PLANT BOARD OF FLORIDA, GAINESVILLE, FLORIDA

THE RENIFORM AND STING NEMATODES IN ALABAMA

Norman A. Minton[1] and Bruce E. Hopper[2]

The reniform nematode, Rotylenchulus reniformis Linford and Oliveira, 1940 was found in a cotton field in eastern Alabama (Chambers County) in 1957. In the course of a survey, soil and roots of cotton were collected from a field in which cotton had made poor growth. Examination of the soil revealed the presence of many larvae and males. However, no adult females were recovered from the roots. To verify the infestation by obtaining mature females, soil was collected from the same field, placed in pots in the greenhouse, and planted to Rowden variety of cotton and to cucumbers. Within 4 weeks, adult females of Rotylenchulus reniformis were recovered from the roots of both species of plants.

The sting nematode, Belonolaimus longicaudatus Rau, 1958 was found in cotton fields in three different locations in Bullock County and in one location with peanuts in Henry County in southeastern Alabama in 1957. Previous records of Belonolaimus[3] indicated that its occurrence was suspected in several locations in Alabama, although its existence was never confirmed.

The reniform and sting nematodes are known to be of importance as root parasites of cotton and several other hosts. The survey indicated they are not as widespread as the root-knot nematodes in Alabama.

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[3]Holdeman, Q. L. 1955. The present known distribution of the sting nematode, Belonolaimus gracilis, in the coastal plain of the southeastern United States. Plant Disease Reptr. 39: 5-8.

INCREASE OF BELONOLAIMUS LONGICAUDATUS ON VARIOUS PLANT SPECIES IN ARTIFICIALLY INOCULATED SOIL[1]

W. H. Lautz[2]

Summary

One to 3 horticultural varieties of 11 crop plant species, 2 grasses, and 1 weed plant in 6-inch and 8-inch pots were inoculated with 10 to 40 picked specimens of sting nematode, Belonolaimus longicaudatus Rau, 1958. Population-increase indices, after 33 to 119 days, ranged from 0 with Bidens sp. (sticktight) to 46 with potato and 125 with soybean. Intermediate indices were obtained with cabbage, cantaloupe, snap bean, field corn, Bermuda grass, and ryegrass. Indices of squash, carrot, cucumber, peanut, and pepper were low.

According to recently published information (6) the common sting nematode in the vicinity of Sanford, Florida is Belonolaimus longicaudatus Rau, 1958. This nematode may or may not be that referred to as B. gracilis Steiner, 1949 in host tests and host lists previously published (1, 3, 4, 5). To obtain information on increase of B. longicaudatus on certain plants, which is fundamental in planning rotations, a series of experiments were conducted during 1957 and 1958.

METHODS

Experiments were conducted in 6-inch or 8-inch pots in the greenhouse. All soil used was Leon fine sand which had been treated with methyl bromide to kill nematodes and other contaminating organisms. Potatoes and corn were planted directly into the pots. All other plants were started in flats and transplanted into the pots after 2 to 4 weeks. There was one plant per pot, with the exception of three plants for ryegrass. Sting nematodes for inoculum were obtained from naturally infested turf and field soil in the vicinity of Sanford, Florida, and were identified and picked out by George J. Rau as Belonolaimus longicaudatus. Nematodes were extracted from the soil by sieving and Baermann funnels, and mature females were picked individually into small quantities of water. The plants were inoculated by pouring the water into small holes punched in the soil around the roots of the plants. A week after the plants were transplanted 5-5-7 fertilizer was applied. At monthly intervals after inoculation the entire contents of individual pots were processed to extract the nematodes by the method of Christie and Perry (2), by using 28- and 200-mesh sieves for the washing and 44-micron nylon screens in the Baermann funnels.

RESULTS

The quantities of plant-parasitic nematodes (Table 1) are the maximum numbers counted in 2 to 4 monthly observations. The maxima with most species were reached at the second observation, but with soybean at the fourth and last observation. Numbers observed were highly variable even in pots inoculated with the same number of nematodes on the same day and examined at the same time. In tests using less than 10 nematodes for inoculum, there were numerous pots in which no increase was observed even with plant species showing high increases when inoculated with larger numbers. Of 49 uninoculated check pots containing the plant species listed in the table, only two were found to contain sting nematodes when the soil was examined.

Increase index figures were calculated by dividing the maximum number of nematodes

[1]Cooperative investigations of the Crops Research Division, Agricultural Research Service, United States Department of Agriculture and the Central Florida Experiment Station.
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Table 1. Maximum quantities of sting nematodes observed in 6- or 8-inch pots containing various plant species.

Species	Horticultural variety and common name	Number of days after inoculation observed	Number of sting nematodes: Used as inoculum	Observed	Increase index
Arachis hypogaea	Spanish peanut	37	20	110	5.5
Bidens sp.	sticktight	43	20	0	0
		43	40	0	0
Brassica oleracea	Late Flat Dutch cabbage	77	10	101	10.1
Capsicum frutescens	Yolo Wonder pepper	33	10	4	0.4
Cucumis melo	Powdery Mildew Resistant 45 cantaloupe	89	10	147	14.7
Cucumis sativus	Early Surecrop hybrid cucumber	35	10	2	0.2
Cucurbita pepo	Yellow Summer Crookneck squash	77	10	25	2.5
Cynodon dactylon	common Bermuda grass	62	10	111	11.1
Daucus carota	Imperator carrot	65	10	10	1.0
		42	20	12	0.6
Glycine max	CNS 24 soybean	119	20	2496	124.8
		98	40	4620	115.5
Lolium multiflourm	Italian ryegrass	62	10	211	21.1
Phaseolus vulgaris	Tendergreen snap bean	69	20	95	4.8
		60	40	412	10.3
Solanum tuberosum	Irish Cobbler potato	98	40	1840	46.0
Zea mays	Dixie Hybrid 18 field corn	47	10	244	24.4

found by the number used for inoculation. These can be put into three distinct groups as follows:

1. High increase indices (46 to 125), observed in pots containing potato and soybean.
2. Low increase indices (2.5 to 24), observed in pots containing cantaloupe and field corn.
3. No increase, observed in pots containing carrots and pepper.

Plants in the first group apparently are good hosts of B. longicaudatus, those in the second group poor hosts, and those in the third group not hosts.

There was also considerable variation in the time at which the maximum population was observed, but this had no constant relation to the maximum numbers observed.

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THE RELATION OF RAINFALL, RELATIVE HUMIDITY, AND TEMPERATURE TO LATE BLIGHT IN MAINE

R. A. Hyre, Reiner Bonde, and Barbara Johnson[1]

Abstract

The first observation of blight, due to Phytophthora infestans, and the estimated mean foliage blight of about 7 percent were quite accurately reflected by the three methods investigated for predicting blight. One method was based on rainfall and temperature data, the other two on relative humidity and temperature data.

INTRODUCTION

A study was continued in 1958 of the relation of rainfall, relative humidity, and temperature to the occurrence of late blight of potato (due to Phytophthora infestans Mont. D By) in Aroostook County, Maine. The instrumentation included, for the first time, an Auchincruive self-calculating blight forecast recorder. At the end of the season the amount of foliage blight in the county was estimated. During the season a blight forecast and warning service was conducted by the junior authors.

METHODS

Three methods were used for resolving the weather data. The first was similar to that used in Maine in 1957 (1). Rainfall and temperature data were obtained from collaborators of the United States Weather Bureau for the four stations of Fort Kent, Caribou, Presque Isle, and Houlton. The initial occurrence of blight was forecast after 10 consecutive days when both rainfall and temperature were favorable and when the current weather forecast was for continued blight-favorable weather. Rainfall was considered favorable when the 10-day total was 1.20 inches or more. Temperature was considered favorable when the 5-day average was less than 78° F. Any day was considered unfavorable, however, if the minimum temperature was less than 45° F. An unfavorable day due to low temperature was not allowed to interrupt the count of consecutive favorable days. That day was simply omitted from the count. The disease was anticipated 1 or 2 weeks after it was forecast. Once blight was established 10 favorable days were no longer required for it to spread.

With the second and third methods blight favorable periods were determined from relative humidity and temperature data. These records were obtained in 1958 by placing a hygrothermograph and an Auchincruive self-calculating blight forecast recorder 4 feet above ground at Aroostook Farm, Presque Isle, in a standard Weather Bureau shelter. With the 90 percent humidity method (Wallin, Jack R. U.S. Dept. Agr. Plant Disease Situation Report 28, 1957) favorable periods were rated for blight severity as follows:

Temperature range° F[a]	Severity value			
	1 (trace)	2 (slight)	3 (moderate	4 (severe)
	Time in hours			
45 - 53	16 - 18	19 - 21	22 - 24	25 -
54 - 59	13 - 15	16 - 18	19 - 21	22 -
60 - 80	10 - 12	13 - 15	16 - 18	19 -

[a] The mean temperature of the period when relative humidity ≥ 90 percent.

A tentative interpretation of severity value was as follows: a trace of blight was expected with a severity value of one per week, slight with a value of two per week, etc.

The hygrothermograph was put in place at the time of emergence of the potatoes and a cumulated severity value of about 20 was suggested[2] as a "zero" time -- before which blight

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[2] Personal communication from Jack R. Wallin.

would not be forecast. These criteria were developed, however, for an instrument placed 12 to 15 inches above the ground among the vines.

The 75 percent humidity method was used to determine "Beaumont periods." Such periods occurred when the relative humidity was at or above 75 percent and the temperature was at or above 50° F for 48 consecutive hours. In England a zero time is involved and until that date is reached Beaumont periods are not considered valid (2). The concept of a zero date is recognition of a period of time required for the fungus to become established and in a position to cause an epiphytotic. Such a concept is implicit, also, in the 10 consecutive favorable days required for a positive forecast of initial blight by the rainfall temperature method.

The determination of Beaumont periods is simplified by the use of an Auchincruive self-calculating blight forecast recorder (2) shown in Figure 1. It is so constructed that the mark

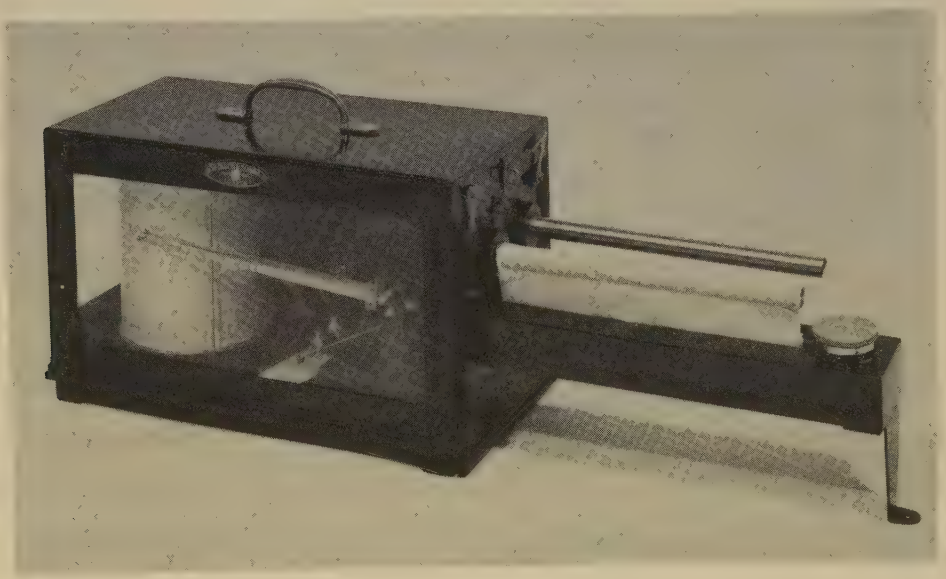

FIGURE 1. An Auchincruive self-calculating blight forecast recorder.

of the wet bulb system is on or above the mark of the dry bulb system at any temperature when the relative humidity is at or above 75 percent.

Unfortunately, progress curves for the development of blight were not obtained; but, during the first week of September, the authors surveyed the county for the amount of foliage blight present at that time. The key used was that adopted in England for use by the National Agricultural Advisory Service and the National Institute of Agricultural Botany (3).

RESULTS

Figure 2 shows an analysis of rainfall, relative humidity, and temperature data for Fort Kent, Caribou, Presque Isle, and Houlton as related to the occurrence of blight. The amount of foliage blight present in early September is given in Table 1.

Table 1. Amount of potato late blight infection in Aroostook County, Maine, as estimated September 3 - 6, 1958.

Location	Number of fields	Percent foliage blight	
		Range	Average
Northern Aroostook (Fort Kent)	69	0-95	19.8
Central Aroostook (Caribou and Presque Isle)	141	0-95	4.2
Southern Aroostook (Houlton)	83	0-75	2.3
Total	293		7.3

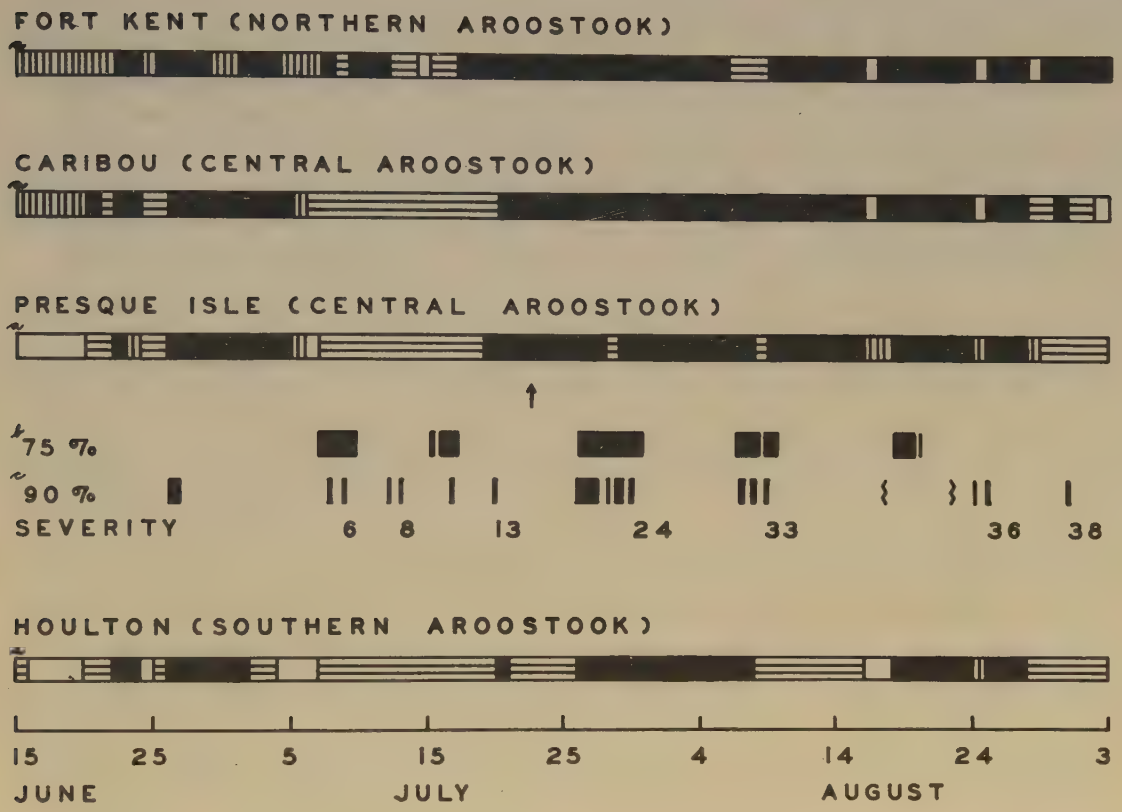

FIGURE 2. Three methods of analyzing weather data in relation to the occurrence of late blight of potato in Aroostook County, Maine, in 1958.

The season of 1958 was very favorable for late blight. Dr. Schultz, United States Department of Agriculture, found late blight on two dump piles near Presque Isle on June 3 -- the earliest it was ever found on dump piles in Aroostook County. In the fields the potato sprouts had not yet emerged on that date. The disease was widespread on dump piles this year. On commercial potatoes the disease was first found on July 22 in three fields in the vicinity of infected dump piles in central Aroostook County. Stalk lesions were 3 to 4 inches long at that time. Infection was estimated to have occurred 10 to 15 days earlier. This early infection apparently occurred soon after a period of 9 or 10 consecutive blight-favorable days in late June and early July (see Figure 2). Also, it was about the time of the first Beaumont period of 65 hours registered by the 75 percent humidity method and about the time of a cumulated severity value of 6 to 8 by the 90 percent humidity method. Later weather was indicated to be favorable for blight by all three methods and the disease increased rapidly. In early September the estimated foliage blight was over 7 percent for the county with the least amount of blight in southern Aroostook where the weather was less favorable for the disease.

The first observance of blight and its later increase would seem to have been quite accurately reflected by the rainfall-temperature method. The fact that more blight was not present is a tribute to the control program of most growers since the 1958 season was very favorable for the disease. For the period of June 4 to September 3 rainfall occurred on 30 percent of the days at Houlton and on 55 percent of the days at Fort Kent with a total of 18.66 inches at the latter place. In spite of the excessive rainfall 50 percent of the fields sampled in the county had 0.1 percent or less of foliage blight in early September. No blight was found in 20 percent of the fields but about 9 percent had 50 to 95 percent of the foliage blighted. The amount of blight present in different parts of the county tended to vary according to the number of blight-favorable days indicated by the forecast method but the effect of the favorable days was modified considerably by the degree of control obtained by different growers.

The field infections apparently occurred about the time of the first Beaumont period of the season recorded by the Auchincruive self-calculating blight forecast recorder. Further experience with this machine in Maine will be necessary, however, to determine its value for forecasting blight. By the 90 percent humidity method there was a cumulative severity value of about 6 to 8 at the time of estimated first field infections. There were later infection periods to correspond with the increase in the amount of blight.

The present study indicates that a degree of flexibility is desirable in applying the criteria for determining favorable blight periods. With the rainfall-temperature technique this last season, for instance, the favorable days required may not have been strictly consecutive; 9 or possibly 8 days may have been sufficient, or a 10-day total rainfall needed may have been 1.10 inches instead of 1.20 inches. The second, fourth, and fifth Beaumont periods listed had a break of 2 or 3 hours when the relative humidity was slightly under 75 percent. Their inclusion, however, would seem to better indicate the amount of blight that developed. The use of a degree of flexibility in applying the criteria comes with experience and skill and does not impair the usefulness of the methods for the purpose for which they were developed.

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PROGRESS REPORT OF NATIONAL SCREENING COMMITTEE FOR DISEASE RESISTANCE IN THE TOMATO FOR 1954-1957[1]

Summarized by Leonard J. Alexander[2]

PRELIMINARY REPORT ON SCREENING WILD SPECIES FOR DISEASE RESISTANCE

Most of the wild species were distributed in the first lot of accessions for disease resistant screening. This work was summarized by Alexander and Hoover and published in 1953 (1) and 1955 (2). Since then two additional lots of accessions have been distributed to the cooperators for screening. These include a few of the wild species but mostly are accessions of the type species Lycopersicon esculentum and Suspected natural crosses L. esculentum x L. pimpinellifolium. The former is separated into two groups: type species including f. pyriforme and var. cerasiforme, and named and presumed varieties.

This report is in summary form and concerns the second and third lots of accessions distributed for screening. It is planned to publish the screening reports in more detail later. However, this summary will allow investigators to immediately make use of the data now at hand.

Twenty-nine cooperators in 11 States, the United States Department of Agriculture, Canada, Jersey of the Channel Islands and two commercial companies took part in this screening program for disease resistance to 15 diseases. The work contributed by each cooperator is shown in the discussion of each disease.

It has been pointed out by several cooperators engaged in this evaluation for disease resistance that the accessions as distributed were not homozygous. None of the accessions used in this work has been pure lined. They are in the same condition as received from the collectors and in many cases are heterozygous. It was thought unwise to attempt to make selections when seed was multiplied, because of the danger of losing some valuable germ plasm. Rather, it was felt that the accessions should be distributed in the condition in which they were received and if a research worker found a particular gene for resistance, suited to his needs, he could then pure line the accession for that character. One exception to this procedure was followed, that is, in those few cases where obvious mixtures were present no seed was saved from the contaminants. The tomato accessions were described by Hoover et al., 1955 (3).

Tomato accession numbers referred to in this report are those assigned to the tomato stocks by the New Crops Research Branch[3], Crops Research Division, Agricultural Research Service, United States Department of Agriculture. Two sets of accessions of 150 each, referred to as the second and third lots, have been distributed for classification for disease resistance. In those cases where the cooperators have reported on more than one set of accessions, they will be summarized separately in this bulletin. In other cases where only one set of accessions has been reported on, the report will cover only that part of the screening program. The separate reports from each disease are listed in alphabetical order.

Accessions were distributed to the various cooperators from the Regional Plant Introduction Station at Ames, Iowa by Dr. M. M. Hoover and his successor, Dr. W. H. Skrdla. The classification and numbers of the second and third lots of accessions are given in the two Plant Introductory lists that follow.

ALTERNARIA LEAF SPOT AND COLLAR ROT (ALTERNARIA SOLANI)

Reports on screening for resistance to Alternaria solani were submitted by Andrus, Harrison, Linn, Porte, and Wingard. Reports of these collaborators were summarized by Andrus. The reports of the different collaborators are in conflict, if the early blight and collar rot phases are taken together as a single disease. However, if resistance to the two phases, early blight and collar rot, are considered separately, then the results of the screening work appear to give some evidence that resistance exists. So far no complete immunity has been

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[1] Published with the approval of the Director of the Ohio Agricultural Experiment Station as Journal Article No. 85-58.

[2] Professor of Plant Pathology, Ohio Agricultural Experiment Station, Wooster, Ohio.

[3] Formerly the Plant Introduction Section, Horticultural Research Branch, Agricultural Research Service.

Plant Introductory numbers by species for the second lot of accessions distributed for disease resistant screening.

L. pimpinellifolium	205011	109832	128218	163245	165053	169573
205009	205013	109838	128219	163247	165489	169574
205012	205015	110596	128220	163248	165490	169575
205014	205016	110597	128231	163249	166985	169576
Species Crosses (L. esculentum x L. pimpinellifolium)	205017	110946	128236	163251	166986	169577
198912	205018	114490	128240	163252	166989	169579
204975	205019	115201	128244	163253	166991	169580
204976	205020	117222	128246	163254	167041	169581
204978	205021	117564	128247	163255	167074	169582
204980	205022	117565	128249	164177	167099	169583
204981	205026	118408	128272	164278	167103	169584
204982	205033	119776	128280	164290	167141	169586
204987	L. esculentum:	121662	128281	164478	167206	169587
204992	type species incl. f. pyriforme and	124034	129052	164482	167329	169588
204994	var. cerasiforme 91916	124036	129071	164541	169565	169589
204995	92356	124161	129082	164628	169566	169590
204996	92853	124165	129098	164673	169567	171708
204997	92863	126913	129111	164719	169568	171709
204998	95585	126918	159181	164945	169569	171710
204999	95587	127815	159193	164946	169570	171711
205002	97321	127820	159198	164947	169571	171716
205010	103055	128174	162679	165030	169572	171717

discovered in this survey. Dr. Andrus feels that we have the same level of resistance, found in this survey, already incorporated into some commercial breeding stocks. The results of the screening of the second group of accessions are reported as follows:

Alternaria reaction of tomato accessions screened in 1953-1954:

Relative resistance to both leaf and stem phases

L. peruvianum 128646 L. pimpinellifolium 205012

Suspected crosses

(L. esculentum 204992 x L. pimpinellifolium 205019)

Relative resistance to leaf phase but susceptible to stem phase:

L. hirsutum 126445 127827 L. pimpinellifolium 126432 205014

Plant Introductory numbers by species for the third lot of accessions distributed for disease resistant screening.

L. peruvianum
126928
212407

L. pimpinelli-folium
211838
211839
211840
212408
212409

Suspected cross (*L. esculentum* x *L. pimpinelli-folium*)
126951
129024
158161
158171
190188
195003
195006
195322
195324
195325
195788
195789
195790
195791
197159
204587
209975
212410
212411

L. esculentum type species incl. f. *pyri-forme* and var. *cerasiforme*
65023
91908
91909
91911
91912
91913
91914
91917
91919
92854
92855
92856
92857
92859
92860
92861
92864
92865
92866
93302
95583
95584
95588
95589
95590
95592
97538
99782
100697
102714
102715
102716
102717
102719
102721
102722
102724
102725
102884
102885
102886
105225
105266
105267
105342

L. esculentum Named and presumed varieties)
91458
91907
91918
95591
98097
102713
106997
109113
109315
109514
109831
109833
109834
109835
108244
108246
109112
109316
109512
114966
116219
118326
118406
118685
116526
118328
118686
121663
121664
121665
121666
121667
123433
123434
123435
123436
123437
123438
127802
127808
109836
109837
111406
111407
111408
111409
113516
115219
115599
115872
128214
128285
128288
128291
128292
128293
128338
128586
128589
128591
128592
128597
128606
128886
128887
128888
128890
129043
129113
129126
129132
129138
129139
129690
135909
136475
140050
141963
146090

Type species		Suspected crosses	
L. esculentum		(L. esculentum x L. pimpinellifolium)	
127815	164719	112835	205002
		118403	205011
		204998	205020

Relative resistance to stem phase but susceptible to leaf phase:

Type species			
L. esculentum		L. pimpinellifolium	
124165	128174	205009	
159193			
		Suspected crosses	
L. glandulosum		(L. esculentum x L. pimpinellifolium)	
126441		204978	205013
		205010	205015

Several accessions had mixed or doubtful reactions. Many accessions were susceptible to both phases.

The results secured from screening the third group of accessions are reported as follows:

Alternaria reaction of tomato accessions screened in 1955-1956:

Relative resistance to both leaf and stem phases

Suspected crosses	
(L. esculentum x L. pimpinellifolium)	
190188	204587
195324	212411

Relative resistance to collar rot but doubtful in reaction to early blight:

		Suspected crosses	
L. peruvianum		(L. esculentum x L. pimpinellifolium)	
126928		158171	195788
		195006	195789
		195325	212410
		Type species	
L. pimpinellifolium		L. esculentum	
211838	212408	97538	109112
211839	212409	105266	109316
211840			

Named and presumed variety

L. esculentum
123436

Relative resistance to collar rot but susceptible to early blight:

Suspected crosses		Type species
(L. esculentum x L. pimpinellifolium)		L. esculentum
195791	197159	108244

Named and presumed varieties

L. esculentum	
106997	111408
111406	121663
111407	128890

Many accessions were susceptible to both phases.

ANTHRACNOSE (COLLETOTRICHUM PHOMOIDES)

Tests for anthracnose on the second lot of seed was reported by Lester Schaible of the Campbell Soup Company and on the third lot of seed by A. D. Hoadley, also of the Campbell Soup Company. In the 1953 and 1954 data accessions listed following were found to possess resistance. Three accessions had a high level of resistance. Four of the accessions in the suspected crosses and 15 of the type species were found to possess an intermediate level of resistance. In the 1955 and 1956 data, nine accessions had a high level of resistance. The results are listed as follows:

Relative resistance to anthracnose in 1953 and 1954:

High level of resistance to anthracnose

Type species

L. esculentum

92863 128244 171708

Intermediate level of resistance to anthracnose:

Suspected crosses

(*L. esculentum* x *L. pimpinellifolium*)

204982 205015

205010 205016

Type species

L. esculentum

128240 129098 169572

128249 159193 169581

129052 163252 169586

129071 163254 169587

129802 166985 204977

Relative resistance to anthracnose in 1955 and 1956:

High level of resistance to anthracnose

Type species

L. esculentum

92855 92859

Named and presumed varieties

(*L. esculentum*)

128292

Suspected crosses

(*L. esculentum* x *L. pimpinellifolium*)

158171 197159

195006 204587

195791 212411

LEAF MOLD (CLADOSPORIUM FULVUM)

A. E. Kerr of the Vineland Experiment Station, Vineland, Ontario, classified both groups of accessions for resistance to tomato leaf mold caused by *Cladosporium fulvum*. He used race 6 of the pathogen for the classification. The results are listed as follows:

Relative resistance to leaf mold in 1953 and 1954:

Intermediate resistance

Type species

L. esculentum

124161

Relative resistance to leaf mold in 1955 and 1956:

High resistance

L. peruvianum

126928 212407

L. pimpinellifolium

211840

Intermediate resistance

L. pimpinellifolium

211838 212408

211839 212409

DIDYMELLA CANKER (DIDYMELLA LYCOPERSICI)

The work to test the accessions for resistance to *Didymella lycopersici* was recently undertaken at the States Experiment Station, Trinity, Jersey, C. I. D. H. Phillips made the tests. He reported that P. I. accessions 126448 and 199380, both species of *L. glandulosum*, were resistant to *Didymella lycopersici*. It is assumed that the other accessions tested were susceptible.

FROST RESISTANCE

Dr. G. A. Kemp, of Letheridge, Alberta, tested the accessions for frost resistance. This is a difficult test to conduct out-of-doors because the temperatures either do not drop sufficiently or, if the plants are planted too early, the temperatures are apt to drop to extremes and kill all the plants. Therefore, the results so far secured have not been encouraging from the standpoint of locating frost resistance.

However, in the fall of 1957, certain accessions of the third lot were exposed, for a short period, to two degrees of frost. Injury varied from no damage to severe. The accessions are listed following which showed little or no damage.

No damage		Very slight damage
	L. peruvianum	212407
	L. pimpinellifolium	211838

No damage		Very slight damage
	Type species (L. esculentum)	
97538		91909
99782		92865
136475		92866
		102717
	Named and presumed variety	
	L. esculentum	123437

FUSARIUM WILT (FUSARIUM OXYSPORUM F. LYCOPERSICI)

Tests for Fusarium wilt resistance were reported by Lesley, Middleton, Porte, Alexander, Harrison, and Epps. Alexander reported on resistance to race 2 in addition to resistance to race 1. The cultures used were those in possession of the individual cooperators. The results are based on seedling greenhouse tests.

Fusarium reaction of tomato accessions to race 1 of the pathogen screened in 1952-1953:

Five reports were received which gave the results of testing the second lot of accessions for resistance to race 1 of the pathogen. One cooperator reported on resistance to race 2. Six accessions of this second lot of 150 tested showed a high degree of resistance to race 1 of the pathogen. The distribution of these accessions is reported as follows:

L. pimpinellifolium	Suspected crosses (L. esculentum x L. pimpinellifolium)		
205012	204987	205026	205033

Named and presumed varieties (L. esculentum)	Type species (L. esculentum)
115201	126913

Accession 205033 appeared to have the highest resistance to race 1. Other accessions had somewhat less resistance. Still others, for example 166991 and 169576, apparently had moderate resistance.

Fusarium reaction of tomato accessions to race 2 of the pathogen screened in 1952-1953:

Only one accession, 205010, Suspected crosses (L. esculentum x L. pimpinellifolium), showed appreciable resistance to race 2 of the pathogen. Three other accessions 117565, 126913, and 159181 showed slight resistance to race 2.

Only one cooperator reported on the third group of accessions. This cooperator reported on reactions to both races 1 and 2.

Fusarium reaction of tomato accessions to race 1 of the pathogen screened in 1955-1957:

High resistance		Intermediate resistance	
L. peruvianum			
126928	212407		
L. pimpinellifolium			
211838	212408	211840	212409
211839			
Suspected crosses (*L. esculentum* x *L. pimpinellifolium*)			
		126951	195006
		195789	195790
Named and presumed varieties (*L. esculentum*)			
		116526	123437
		118686	123438
		123434	128292
		123436	146090
Type species (*L. esculentum*)			
		91908	102714

Fusarium reaction of tomato accession to race 2 of the pathogen tested in 1955-1957:

High resistance			Intermediate resistance		
L. peruvianum					
126928	212407				
L. pimpinellifolium					
211840	212409		211838	211839	
212408					
Suspected crosses (*L. esculentum* x *L. pimpinellifolium*)					
			126951	195790	
Named and presumed varieties (*L. esculentum*)					
			109113	118686	123438
			109514	121663	127802
			109836	121665	128285
			111407	121667	128888
			111408	123433	129138
			111409	123434	146090
			116526	123437	
Types species (*L. esculentum*)					
			92861	99782	105225
			92865	102714	114966

From the reports of these investigators it should be noted that certain accessions exhibit evidence of resistance to both races 1 and 2 of the pathogen. This was especially evident on the third lot of accessions tested in 1955-1957. However, confirmation from other workers is needed.

In the species *L. peruvianum* both accessions 126928 and 212407 appeared to be highly resistant to both races of the pathogen. Also accession 211408, *L. pimpinellifolium*, appeared to be resistant to both races of the fungus. These are worthwhile observations because they indicate that should race 2 become widespread and cause serious losses, a good source of resistance to it is at hand.

PHOMA BLACK-SPOT (PHOMA DESTRUCTIVA)

Accessions were screened for resistance to Phoma black-spot at the Bradenton, Florida Experiment Station by J. M. Walter. Unfortunately the results of the tests were confused by the development of Stemphylium leaf spot. It had previously been reported that accession 115201 had a high level of resistance. This has been confirmed, but Walter reported that it

appears that the resistance of this accession is difficult to handle in a breeding program and it is therefore desirable to continue testing in hopes of finding an accession with a superior type of resistance that can be used readily in a breeding program.

Accessions which may possess a high level of resistance to Phoma black-spot:

Type species (L. esculentum)	
167041	169575
167074	169580
169566	169581
169567	169582
169571	

TOMATO LATE BLIGHT (PHYTOPHTHORA INFESTANS)

The first lot of accessions was screened for resistance to two races of the tomato late blight fungus, Phytophthora infestans, by investigators at the University of West Virginia and at Ottawa, Canada. Graham screened 50 plants of each accession for resistance to his tomato race 0; resistant plants were saved from accessions which had more than 10 individuals surviving. At West Virginia University, Morgantown, Gallegly and Marvel tested approximately 25 plants of each accession with their tomato races 0 and 1.

The data in the Plant Introductory list (second lot) show that all of the accessions in the L. pimpinellifolium and Suspected crosses (L. esculentum x L. pimpinellifolium) groups possess resistance to tomato race 0 except accessions 204975, 205022, and 205033. Some of these accessions were homozygous for resistance while others were heterozygous. The mean disease indices of some accessions suggested that resistance was controlled by a dominant gene (indices of 1 to 2.5) whereas the indices of others suggested that resistance is controlled by multiple genes (indices of 2.6 to 3.9). Breeding behavior is necessary, in most cases, before the mode of inheritance can be stated.

None of the accessions in the L. esculentum group was highly resistant to tomato race 0. Some accessions showed a low to moderate level of multiple-gene resistance.

The results from screening with tomato race 1 show only 3 accessions with resistance. The pattern of segregation and the mean disease indices of these accessions (204996, 205016 and 205017) Suspected crosses (L. esculentum x L. pimpinellifolium) suggested that resistance is controlled by multiple genes. Accession 204996 is considered the most promising among this group as a source of resistance to both races of the tomato late blight fungus.

MOSAIC (POTATO VIRUS Y)

J. M. Walter of the Bradenton, Florida Experiment Station tested the two lots of accessions for resistance to potato virus Y. In the second test none of the accessions was resistant to the virus although there appeared to be several degrees of severity of symptoms. The question of strains of the virus arose, but since information concerning strain reaction on hosts was not available, Walter used a laboratory strain which he had in stock for screening the first two lots of accessions. P. I. accession 126913 exhibited the mildest symptoms of those tested.

Among the third lot of accessions tested, three showed encouraging results. They are 128887, Presumed varieties (L. esculentum), and 195006 and 205019, Suspected crosses (L. esculentum x L. pimpinellifolium). However, before definite conclusions can be drawn, Walter considers it necessary to test with several strains of the virus. Walter also reported on a fourth lot of accessions and on certain selected plants from accessions previously screened. In his last report he tested for resistance to severe and mild strains of the virus. His last report confirms the previous report that accession 128887 is highly resistant. He also found a new accession 117897, Type species (L. esculentum), to be highly resistant. Individual plants from these two accessions were re-inoculated four times. Twenty-five plants of accession 128887 and 26 plants of accession 117897 remained healthy.

ROOT-KNOT NEMATODE (MELOIDOGYNE SP.)

The two lots of accessions were tested by two cooperators, A. L. Taylor and associates, and A. L. Harrison. Harrison alone reported tests on the second lot of accessions. In his

work he used M. incognita, M. incognita var. acrita, M. javanica, M. arenaria, and Meloidogyne sp.

Of the accessions tested in the second lot, none was highly resistant. Six accessions showed moderate resistance to M. javanica. These accessions were in the Suspected crosses (L. esculentum x L. pimpinellifolium) and type species (L. esculentum). The accessions with indices follows.

Type species (L. esculentum)

Accession	Indices
118408	2.2
124036	2.1
127820	3.6
128246	2.6

Suspected crosses
(L. esculentum x L. pimpinellifolium)

Accession	Indices
205002	3.2
205010	2.2

It is to be noted that none of the accessions showed high resistance with an index of one when tested against M. javanica. All accessions were susceptible to all other collections of nematodes.

Both cooperators tested the third lot of accessions for nematode resistance. A. L. Harrison reported on only one nematode reaction. It is assumed that he used the Meloidogyne sp. culture. F. J. Spruyt, working with A. L. Taylor, reported on the reaction of four species of nematodes. These are M. incognita var. acrita, M. javanica, M. arenaria and M. hapla.

Only the two accessions of L. peruvianum showed worthwhile resistance. All the other accessions were susceptible to nematode infection.

The ratings of the two L. peruvianum accessions are as follows:

Reaction of accessions of L. peruvianum to five nematode tests.

Accession	Meloidogyne sp.	M. incognita var. acrita	M. javanica	M. arenaria	M. hapla
126928	2	3	3	3	3
212407	1	3	2	3	3

In this case, Harrison's results indicated a higher resistance than Spruyt found with the four species he used. However, it does not appear that the degree of resistance is very high even to any one species.

SEPTORIA LEAF SPOT (SEPTORIA LYCOPERSICI)

Three cooperators, C. F. Andrus, Mark L. Tomes, and J. C. Horton, reported on Septoria leaf spot resistance. Of the second lot of accessions only five showed any marked resistance. The degree of resistance of those five could not be called more than intermediate as no accession exhibited more than a 3 reaction.

The distribution of the five accessions by species is as follows:

L. pimpinellifolium
205014

Suspected crosses
(L. esculentum x L. pimpinellifolium)
204987 204999

Type species
(L. esculentum)
128218 129071

The third lot of accessions, 1955-1957, was tested by all three cooperators. Again in this group of accessions very little, if any, high resistance was found, although in this case individuals of some accessions did exhibit a type 2 reaction. This could be taken to indicate that some of the accessions were segregating for the type of resistance they possessed.

The distribution of the accessions by species which exhibited some resistance is indicated as follows:

L. peruvianum
126928 212407

Suspected crosses
(L. esculentum x L. pimpinellifolium)
126951

Type species
(L. esculentum)
111406 111407

STEMPHYLIUM BLIGHT (STEMPHYLIUM SOLANI)

Disease resistant classifications for Stemphylium were received from M. L. Tomes and M. B. Linn and P. M. Miller for the second lot of accessions, 1953-1954. Tomes and Hoadley reported on the third lot of accessions, 1955-1957. All used seedling leaf infection and the agreement in classification was excellent. The results for the second lot of accessions are reported as follows.

Reaction of the second lot of accessions screened for resistance to S. solani, 1953-1954.

Species	Accession	Ratings	
		Tomes	Linn and Miller
L. pimpinellifolium	205012	2	4
Suspected crosses			
(L. esculentum x L. pimpinellifolium)	205033	1	2
Type species (L. esculentum)	118408	2	2

Reaction of the third lot of accessions screened for resistance to S. solani, 1955-1957.

Species	Accession	Ratings	
		Tomes	Hoadley
L. peruvianum	126928	2	1
	212407	2	2
L. pimpinellifolium	211838	1-2	2
	211839	1-2	2
	211840	1-2	2
	212408	1-2	2
	212409	1-2	2
Suspected crosses	190188	1-2	3
(L. esculentum x L. pimpinellifolium)	212410	1-2	3
Type species (L. esculentum)	95584	2-4	2
	99782	2	3
	100697	2-4	3
	106997	1-2	2

It is to be noted that none of the accessions was found to possess a 1 reaction by all classifiers. However, certain accessions apparently have a high degree of resistance. These results indicate that the two species L. peruvianum and L. pimpinellifolium have the highest type of resistance.

TOBACCO ETCH (MARMOR ERODENS)

J. M. Walter of the Bradenton Experiment Station, Florida, tested the first group of accessions for resistance to the tobacco etch virus. He found one accession, 166989, Type species L. esculentum, to be resistant after two inoculations and the plants remained symptomless throughout the season. Walter previously reported that accession 183692, Named and presumed varieties, L. esculentum, was resistant. Thus, so far, two accessions have been isolated which are resistant to the tobacco etch virus.

TOBACCO MOSAIC (MARMOR TABACI)

The second lot of accessions was tested for resistance to a green strain, Johnson's type strain, of the tobacco mosaic virus by Doolittle, Alexander, and Soost. Alexander also tested the same accession for resistance to a yellow strain, McKinney's BSY, of the virus. No evidence of resistance to either strain of the virus was found in these accessions.

The third lot of accessions was tested for resistance to the green strain of TMV by Alexander, Doolittle, Soost, and Thornberry. Alexander again tested these accessions for resistance to McKinney's BSY yellow strain of the virus.

Only two accessions, 126928 and 212407, both L. peruvianum, showed positive evidence of resistance. These accessions segregated for resistance and susceptibility in all tests. One investigator reported certain of the other accessions to be segregating for resistance. However, the other investigators did not corroborate those findings. Another cooperator found certain seedlings to be free of symptoms but when these symptomless plants were assayed for the presence of TMV positive reactions were obtained. Thus it is assumed that with the exception of the two L. peruvianum accessions, all others were susceptible.

VERTICILLIUM WILT (VERTICILLIUM ALBO-ATRUM)

O. C. Cannon and G. E. Wooliams reported on resistance to Verticillium wilt in the second lot of accessions, 1953-1954. Certain plants of several accessions did not develop symptoms of wilt when tested but when these apparently resistant plants were progeny tested there was no evidence of resistance. Thus it appears that there was no resistance in this lot of accessions.

SEED MULTIPLICATION

Credit is due certain investigators who kindly multiplied seed of accessions which fruit sparingly, if at all, in the North Central states. Those who had the best success were A. L. Harrison of Texas and W. J. Virgin of the California Packing Corporation.

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OHIO AGRICULTURAL EXPERIMENT STATION, WOOSTER

UNUSUAL OCCURRENCE OF TWO TOMATO DISEASES IN THE STATE OF SÃO PAULO, BRAZIL DURING 1958

Ferdinando Galli[1]

Tomato is one of the most important vegetable crops in the State of São Paulo, Brazil, and is cultivated chiefly in five different areas of that State, from February to October. Various fungus and virus diseases occur every year, varying their importance according to the occurrence of favorable weather conditions. During the 1958 season two diseases were of abnormal occurrence, and are related below.

Bacterial canker (Corynebacterium michiganense (E. F. Sm.) H. L. Jens.): This disease was first observed by the author on a tomato field in Caucaia, near São Paulo City, and in Piracicaba, in October 1956. The disease was reported as occurring in the Vale do Paraiba, in February 1957[2]. During 1957 bacterial canker spread rapidly and during the 1958 season the disease was found in almost all tomato-growing areas of the State.

Statistical data are not available on losses due to the disease, but they may be valued at about 40 percent of the total crop.

Control measures used elsewhere have not been successful in controlling the disease, possibly because of the particular characters of that vegetable crop in the State of São Paulo. Experiments on the control of bacterial canker are being carried on at present.

The rapid spread of the disease cannot be satisfactorily explained. Indeed, occurrence of favorable weather conditions and the use of disease-carrying seeds may explain, at least partially, the epiphytotic nature of bacterial canker.

Oidium: This quite uncommon disease was found in two untreated fields in different areas. In one of them, a single application of karathane eradicated the fungus.

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[1] Professor of Plant Pathology, University of São Paulo College of Agriculture, Piracicaba, S. P.
[2] Andrade, A. C. 1957. Cancro bacteriano em tomateiro. O Biológico 23(2): 36.

CONTROL OF CANTALOUP POWDERY MILDEW IN ARIZONA WITH FUNGICIDES[1]

Robert B. Marlatt, Ross M. Allen, and Robert T. McKittrick[2]

Abstract

Arizona-grown cantaloups were dusted with Karathane and Ovotran and sprayed with cycloheximide and lithium carbonate to control powdery mildew. Observations made during three seasons showed the best results with 1/2 percent Karathane dust applied bi-weekly at 33 pounds per acre. This treatment gave excellent control of powdery mildew, was not phytotoxic, and increased yields of marketable melons.

Three methods of estimating powdery mildew incidence included counting mildew spots on 40 leaves per 56-foot plot, classing 40 leaves as diseased or healthy and similarly classing 100 leaves. The second method was easiest and as accurate as the rest.

Powdery mildew, caused by Erysiphe cichoracearum DC. ex Merat (5), is one of the most important diseases of cantaloups in Arizona. It appears that both races of powdery mildew (3) occur in the Yuma Valley. Sunrise, a cantaloup variety susceptible to both races, was much more thoroughly covered by the fungus than was an adjacent row of Powdery Mildew Resistant No. 45. The latter variety is the one most commonly grown in the State and occasionally is severely infected despite its resistance to race 1. For this reason, the relative effectiveness of various fungicides has been studied.

Godfrey (1) reported good control of cantaloup powdery mildew with hand-duster applications of Karathane, 2-(1-methylheptyl)-4, 6-dinitrophenyl crotonate and isomers, as a 1 percent dust. If dusted plots were adjacent to a source of inoculum, Godfrey found new infections reappearing 2 weeks after dusting. At rates as high as 40 pounds per acre no phytotoxicity was noted. A leaf scorch has been reported, however, on potato, bean and apple resulting from the use of Karathane sprays (6).

1955 EXPERIMENT

Materials and Methods

Karathane as a 1 percent dust, Ovotran (p-chlorophenyl p-chlorobenzenesulfonate) as a 7 1/2 percent dust, and a spray containing 2 ppm cycloheximide, 3-[2-(3, 5-dimethyl-2-oxocyclohexyl) -2-hydroxyethyl] glutarimide, were applied to powdery-mildew susceptible Hales Best cantaloups. Dusts were applied at 80 pounds and the spray at 22 gallons per acre. Pyrophyllite dust was also applied to help distinguish Karathane or Ovotran phytotoxicity from injury due to the dust carriers. Materials were applied four times at approximately 10-day intervals as fruit was reaching maturity. Each treated plot consisted of five 6-foot-wide cantaloup beds 30 feet long. Plots were randomized as a Latin square. A week after the last application, vines were rated for severity of powdery mildew by counting the number of mildew spots on 20 basal leaves from each plot's center bed. Phytotoxicity was evaluated by calculating the percentage of leaves one-half or more dead on the main runners of five plants in the center bed. Ratings of mildew severity and phytotoxicity were evaluated by analysis of variance.

Results

Plots treated with Karathane or Ovotran had significantly less mildew than did the untreated controls. Cycloheximide-treated plots did not show significantly less mildew than controls.

Dusting plots with Karathane or Ovotran resulted in significantly more dead leaves than on vines dusted with pyrophyllite.

The cycloheximide spray did not result in any significant plant injury. Cantaloup tolerance

[1] Arizona Agricultural Experiment Station Technical Paper No. 503.

[2] Respectively, Associate Plant Pathologist, Assistant Plant Pathologist and Assistant in Research.

to cycloheximide had previously been reported by Marlatt (4).

1957 EXPERIMENT

Materials and Methods

Dusts consisted of 1 percent and 1/2 percent Karathane, each applied weekly and bi-weekly at about 33 pounds per acre. Sprays included cycloheximide 2 ppm, cycloheximide 1 ppm and 2 ppm with 1 percent glycerin added, and lithium carbonate, 5 pounds and 10 pounds per 100 gallons of water. Foliage was wet thoroughly with the sprays. Plots were three beds wide and 27 feet long, replicated six times as randomized blocks. Powdery mildew was evaluated and plots compared as in the 1955 experiment.

Results

All plots receiving Karathane showed significantly less mildew than did the controls. Dusting with 1/2 percent concentration bi-weekly was as effective as using 1 percent and weekly applications. No phytotoxicity was apparent.

The cycloheximide and lithium carbonate sprays showed no control of powdery mildew.

1958 EXPERIMENT

Materials and Methods

Karathane was again applied weekly and bi-weekly as a 1/2 and a 1 percent dust. This year's plots were slightly longer, 56 feet. Four weekly and two bi-weekly dustings were applied. A more thorough estimate of mildew incidence was made by using three methods, that is, counting the mildew spots on 40 basal leaves per plot, classing 40 leaves as either healthy or diseased (one or more spots) and classing 100 leaves as healthy or diseased.

As pointed out by Horsfall and Heuberger (2), counting leaf spots is highly recommended because of its objectivity. Classing 100 leaves as diseased or healthy was, however, much faster than counting spots on 40 leaves. Treatments were evaluated by analysis of variance.

Yields of marketable fruit were recorded throughout the season. Western cantaloups are most often sized as 23, 27, 36 or 45 jumbo melons per crate. Generally, sizes 27, 36 and 45 are the most desirable.

Effect of Karathane on Incidence of Powdery Mildew

Findings were similar to those obtained in 1957; all Karathane treatments had significantly less mildew than did the controls. One-half percent Karathane applied bi-weekly was just as effective as 1 percent applied weekly.

Comparison of Mildew Estimations

In the 1957 experiment, placing 20 leaves into two classes of diseased or healthy seemed to give as accurate an estimate of powdery mildew incidence as did counting all of the spots on the 20 leaves.

The same was true in 1958. Classing 40 leaves as diseased or healthy gave as high an F value in analysis of variance as did a similar classing of 100 leaves. Likewise, totaling mildew spots on 40 leaves required much more time but apparently gave no more accurate estimate of mildew incidence.

Effect of Karathane on Melon Yields

Plots dusted bi-weekly with one-half percent Karathane yielded significantly more of the most desirable sizes 27, 36 and 45 combined than did the other treatments. This was noted when an orthogonal comparison was made of this lightest dust application with the others as a group.

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THE EFFECT OF LINDANE ON CUCUMBER YIELDS WHEN USED WITH VARIOUS FUNGICIDES[1]

H. W. Rankin and L. W. Morgan[2]

Cucumbers are an important secondary crop in the Georgia Coastal Plain. The spring acreage is generally planted to pickling varieties, while slicing varieties are grown in the fall. The control of pickleworm, Diphania nitidalis (Stoll), is usually necessary for economical production of both crops.

Comparative fungicide tests have been conducted at the Georgia Coastal Plain Experiment Station, Tifton, Georgia since 1948 for control of downy mildew and other diseases. In the work with cucumbers lindane was used with the various fungicides to reduce insect injury. A few years' observations indicated that in reducing the insect injury factor an insecticide phytotoxicity factor had been introduced. An experiment was designed to determine if this were true.

Dupree et al. (4) found that either lindane or parathion gave good control of the pickleworm. Brooks and Anderson (2) found that under certain conditions benzene hexachloride (1 percent gamma) dust caused severe injury to the new growth of cucumbers and cantaloupes. Crowell and Morrison (3) indicated that all of the chlorinated hydrocarbons are capable of inflicting injury to cucurbits in general under proper conditions. Anderson and Hofmaster (1) stated that benzene hexachloride 0.5 and 1 percent gamma isomer dusts gave excellent control of pickleworm on cucumbers and cantaloupes but caused some injury to the cucumber plants. Stitt and Evanson (5) found that the stand of cucumber seedlings, 14 days after planting, was significantly reduced by the soil application of 2 pounds gamma of benzene hexachloride per acre. All of the findings have been recorded as injury to the plants.

METHODS AND MATERIALS

The experimental plots were laid out in a plan of six randomized blocks, each block including the treatments and dosages as shown in Table 1. Each plot consisted of four rows, each 30 feet long, and were planted the last week in March. Fertilization and cultivation were normal for the crop. The sprays were applied at weekly intervals and were started a few days

Table 1. The effect of lindane on cucumber yield when used with various fungicides.

Treatment	Amount per 100 gallons spray		Average replicate yield (number)
	Fungicide (pounds)	25 percent lindane (pounds)	
Tribasic Copper	3	0	201
Tribasic Copper + lindane	3	1	159
Ziram	2	0	216
Ziram + lindane	2	1	177
Zineb	1 1/2	0	212
Zineb + lindane	1 1/2	1	182
Maneb	1 1/2	0	204
Maneb + lindane	1 1/2	1	173
Cop-O-Zinc	3	0	219
Cop-O-Zinc + lindane	3	1	160
Lindane alone	0	1	142
Check -- no treatment	0	0	209
LSD at .05 percent level			40
LSD at .01 percent level			53

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before the first picking. They were applied with a small power sprayer that developed 250 pounds' pressure. All fungicides and the insecticide were in the form of wettable powder. Neither downy mildew nor pickleworm were significant factors in yield. The plots were picked six times and data were taken from the two inner rows of each. Yield was used as a measure of phytotoxicity. The data are shown in Table 1 as the total number of cucumbers in all replicates by treatments.

RESULTS

An analysis of variance was run on the above data and on the basis of this the following statements can be made:

When copper fungicides (Tribasic Copper and Cop-O-Zinc) were used there was a significant difference at the 5 percent level in each case between the fungicide and the fungicide plus lindane.

When carbamates were used there was no significant difference, at the 5 percent level, in any single case between the carbamate fungicide and the same fungicide plus lindane. However, when the carbamates are considered as a group the difference between them with and without lindane is significant.

The difference between the untreated check and any fungicide alone is not significant.

The data indicate that when lindane is used on cucumbers it should be combined with a fungicide, preferably a carbamate. It is suggested that fungicides, especially the carbamates, may act as safeners for lindane on this crop.

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STREPTOMYCIN FORMULATIONS AND THEIR RELATION TO THE CONCENTRATION OF THE ANTIBIOTIC IN THE CELL SAP OF VARIOUS PLANTS UNDER DIFFERENT CONDITIONS

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Abstract

In the presence of water the release of streptomycin from Nuclay occurs over a longer period of time than it does from either calcium carbonate or gypsum formulation. Streptomycin can be demonstrated in the cell sap of treated plants within 5 minutes after application and diminishes gradually after 4 to 5 days. Wilting of the plants or low temperatures prevailing at or after the treatment do not interfere with absorption of streptomycin by the plants.

At the present time streptomycin is accepted as reliable for use against many bacterial diseases of plants and a few species of downy mildew fungi. It is being used in both wettable and dust formulations. The apparent preference among growers for wettable types of streptomycin may be due to the fact that the majority of users do not have suitable dusting equipment and to lack of adequate information on the comparative merits of dust versus liquid formulations.

The purpose of this paper is to present experimental data showing the concentration of streptomycin in plant cells obtained when different formulations of the antibiotic are applied to the plants, and the effects of light, darkness, moisture, temperature, and other factors on the level of the antibiotic in the plant juice.

MATERIALS AND METHODS

Cucumber plants (Marketer variety) used in the experiments were grown in the greenhouse to the fourth-leaf stage. To compare absorption of streptomycin by other species of plants the following were used in the seedling stage: pinto bean, almond, apricot, cherry, and peach. Determination of the quantity of streptomycin in plants was made from the juice expressed by the use of a special modified garlic press from Holland after the plants were washed with water, immersed in distilled water for 15 to 20 minutes, rinsed and surface dried with a slightly absorbent high quality tissue paper. Filter paper discs (S & S #740E) were saturated with the extracted plant juice and dried for 60 minutes at 40°C. Agar plates were flooded either with a suspension of a culture of the streptomycin-dependent strain of Escherichia coli or a young culture of Erwinia amylovora and dried for 20 minutes at 40°C. The turbidity for the E. coli suspension was 25 and for E. amylovora 35 on a Klett-Summerson colorimeter.

Properly prepared discs were placed on the seeded plates and read for inhibition zones with an antibiotic zone reader after 24 and 48 hours of incubation at 28°C. All measurements were expressed in mm of diameter and represented the average of at least four readings. The experiments were replicated at least two times.

Dust formulations were prepared by mixing an agricultural grade of a 56 percent streptomycin sulfate, kindly supplied by Merck, Sharp and Dohme Co., with various carriers such as Nuclay and calcium carbonate. After a careful and thorough mixing in a pyrex mortar, the dusts were run through a Wiley mill six times to provide uniformity of a given batch.

To determine release of streptomycin from prepared dusts (in vitro) in water, a sample containing the amount of dust necessary to make a theoretical concentration of streptomycin of 100 ppm was added to 10 ml of water, shaken well for 3 minutes, and centrifuged at high speed in a Serval centrifuge (about 10,000 rpm) for 20 minutes. This provided a very clear solution for subsequent plate tests.

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EXPERIMENTAL RESULTS

Release of Streptomycin in Vitro

The rate of release of streptomycin from successive aqueous extractions was more gradual from Nuclay than from calcium carbonate (Table 1) or dust formulations with the agricultural grade of gypsum (Table 2).

Table 1. Release of streptomycin from successive extractions of a Nuclay and a calcium carbonate formulation containing 1000 ppm of the antibiotic. Test organism, Erwinia amylovora. Diameter of disc, 13 mm (S & S, #740E).

Extraction	Nuclay streptomycin dust Diameter of clear zone	Calcium carbonate streptomycin dust Diameter of clear zone
1st	20.5	24.6
2nd	17.0	21.0
3rd	16.0	18.7
4th	16.8	17.3
5th	15.3	16.1
6th	15.0	13.0
7th	14.3	13.0
8th	14.7	13.0
9th	14.3	13.0
10th	13.0	13.0

Table 2. Release of streptomycin from successive aqueous extractions of a Nuclay and a gypsum formulation (1000 ppm). Test organism, Erwinia amylovora. Diameter of disc, 13 mm (S & S, #740E).

Extraction	Nuclay streptomycin dust Diameter of clear zone	Gypsum streptomycin dust Diameter of clear zone
1st	20.0	24.0
2nd	16.2	21.1
3rd	15.0	20.0
4th	15.9	18.2
5th	15.0	17.1
6th	15.0	14.5
7th	14.3	13.0
8th	14.6	13.0
9th	14.3	13.0
10th	13.0	13.0

The Nuclay and gypsum dust formulations containing 1000 ppm streptomycin were dusted gently on both sides of the leaves of greenhouse-grown cucumber plants that had been slightly dampened by a very fine mist. After predetermined periods of time the dusted leaves were harvested for laboratory bioassaying. The experimental plants were held in a greenhouse

where the temperature fluctuated between 65° and 80°F and the relative humidity was about 80 percent. A typical test (Table 3) shows that streptomycin can be demonstrated in the plant juice in about 5 minutes after it is deposited on the leaves, and reaches a maximum in 1 day.

Table 3. Concentration of streptomycin in plant juice of cucumber leaves dusted with three different formulations of streptomycin and bioassayed from 5 minutes to 24 hours after the treatment. Inhibition zones in mm-dia. Test organism, Erwinia amylovora. Size of disc, 13 mm (S&S, #740E).

Duration of dust on plant	Diameter of inhibition zones in mm		
	Streptomycin (1000 ppm in Nuclay)	Streptomycin (1000 ppm in Nuclay (50%) + gypsum (50%))	Streptomycin (1000 ppm in gypsum)
5 minutes	15	17.2	19.0
10 "	15.4	18.9	19.6
15 "	18.0	19.3	20.0
30 "	18.7	21.5	23.4
60 "	18.5	21.5	23.4
2 hours	20.0	21.6	24.1
24 "	20.8	21.6	24.8

Table 4. Persistence of streptomycin in the juice of greenhouse-grown cucumber plants treated with both wettable and dust formulations of streptomycin. Test organism, Erwinia amylovora. Inhibition zone in mm-dia.

Samples taken after (days)	Diameter of inhibition zones in mm	
	Streptomycin spray (100 ppm)	Streptomycin (1000 ppm dust in Nuclay carrier)
1	18.5	20.0
2	18.4	20.0
3	18.3	20.0
4	18.0	19.8
5	17.2	19.6
6	17.3	19.2
7	17.1	19.0
12	16.7	19.0
14	14.8	16.9

Although the final fate of streptomycin in plant juice is not known, its level in the plant does not appear to remain the same. There seems to be a gradual disappearance of the antibiotic as determined by existing bioassaying techniques. The streptomycin level in the experimental cucumber leaves that were sprayed and dusted with streptomycin formulations began to decrease very slowly about the third day after application but was fairly high even after 2 weeks (Table 4).

Table 5. Content of streptomycin in cucumber juice of both wilted and normal plants treated with the Nuclay-streptomycin (1000 ppm) dust and expressed as inhibition zones (mm-dia.) from bioassaying discs (13 mm S & S, #740E). Test organism, Erwinia amylovora.

Samples taken after (hours)	Inhibition zones in mm	
	Wilted	Normal
1	20.0	20.9
2	20.0	20.8
24	20.3	20.7

To determine the effect of wilting on the absorption of streptomycin, the cucumber plants were allowed to develop a medium degree of wilting by drying the pots in which they were growing. They were then dusted with the Nuclay-streptomycin preparation, as in the preceding experiments. As controls, normal, unwilted plants were similarly dusted. There was no appreciable difference (Table 5) in the amount of streptomycin absorbed in the two series.

Since plant disease protectants are applied at various temperatures, depending on climatic conditions prevailing at the time, it was of interest to determine the effect of temperature on degree of streptomycin penetration into the plant. For this purpose young greenhouse-grown plants of uniform size were sprayed and dusted with streptomycin preparation, held under bell jars at the following temperatures: a) fluctuating between 65° and 80°F, b) 15°C, c) 5°C and d) 2 1/2°C, and bioassayed by the same method as in the previous tests. The results (Table 6), representing averages from several replications, show that temperature had little effect on absorption.

Comparative amount of streptomycin from spray and dust formulations penetrating into the woody and herbaceous plants

Since dust formulations present certain advantages over wettable ones, it is important to know how much of the antibiotic is available when this type of formulation is used on woody and herbaceous plants as measured by penetration into the plant. Apricot, almond, peach, and cherry represented the woody plants tested, and pinto bean and tomato the herbaceous plants. These were sprayed and dusted following the procedure outlined above. All samples collected for the bioassaying work were washed thoroughly in distilled water and frozen before juice was expressed from the leaves (Table 7). Bioassaying of the woody species was done with a streptomycin-dependent strain of Escherichia coli, and of the herbaceous species with Erwinia amylovora. It appears (Table 7) that a considerable quantity of streptomycin can be demonstrated in the plant juice of all the plants studied, regardless of type of formulation, although in many instances the dust formulations gave indication of larger amounts of streptomycin going into the cells when applied on moist leaf surfaces. Penetration of leaves of woody species was as good as in leaves of herbaceous species, although E. amylovora yielded smaller inhibition zones.

DISCUSSION

The question of whether streptomycin can successfully be used in dust formulations is of practical importance. The tendency on the part of the many users of plant protectants was to stay away from this kind of formulation on the ground that sprays seemingly gave a better performance in the past. It is assumed that more plant area is covered by sprays and so more toxicant is placed on the plant and, therefore, a larger surface area is guarded against plant

Table 6. Concentration of streptomycin in juice of greenhouse-grown cucumber plants sprayed with 500 and 1000 ppm streptomycin solutions and dusted with a Nuclay-streptomycin (1000 ppm) dust. Plants were held in a humid atmosphere at greenhouse temperatures, 65° to 80°F, 15°C, 5°C, and 2 1/2°C. Bioassayed with a streptomycin-dependent strain of Escherichia coli. Turbidity: 45 of the Klett-Summerson colorimeter. Two series, a and b, are represented.

	I				II				III				IV			
	Streptomycin formulation															
Temperature	Spray 500 ppm		Spray 1000 ppm		Dust 1000 ppm		Spray 500 ppm		Spray 1000 ppm		Dust 1000 ppm		Spray 500 ppm		Spray 1000 ppm		Dust 1000 ppm	
	Diameter of inhibition zones in mm															
	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b
65° to 80°F	25.8	26.8	26.8	28.2	24.9	25.9	25.1	23.5	25.1	28.6	27.0	24.1	18.5	20.1	21.9	25.8	18.0	21.1	16.9	20.1	28.6	25.2	19.0	18.7
15°C	24.0	26.0	26.7	29.0	23.6	23.1	24.4	25.7	25.4	28.2	26.8	28.7	18.6	22.2	21.0	26.7	15.8	16.2	28.6	24.7	24.4	25.9	20.5	20.7
5°C	23.2	26.3	24.2	28.1	27.2	26.4	25.7	26.2	26.5	25.6	26.9	29.1	20.1	20.9	22.0	24.1	26.4	27.9	23.0	26.5	23.6	27.1	21.5	24.6
2 1/2°C	24.5	25.1	28.1	26.7	26.7	30.4	25.5	21.0	26.7	25.9	29.7	31.3	20.4	23.8	21.6	23.1	24.3	23.4	20.2	26.4	22.4	26.8	22.8	25.7

Table 7. Availability of streptomycin from sprays and dusts when applied on woody (almond, apricot, cherry, pear) plants versus herbaceous plants (pinto bean, and tomato). Plants were held on a greenhouse bench at 65° to 80°F. Relative humidity in the greenhouse between 70 and 80 percent. Test organisms, Escherichia coli for woody and Erwinia amylovora for herbaceous plants. Bacterial growth in the case of E. coli and inhibition zones expressed in mm of diameter. Discs used, S & S, #740E (13 mm dia).

Species of plants	Streptomycin formulation Diameter of inhibition zones in mm	
	Streptomycin dust (2000 ppm)	Streptomycin spray (1000 ppm)
Almond, Prunus amygdalus L.	46	43
Apricot, P. armeniacum L.	42	45
Cherry, P. avium L.	40	32
Peach, P. persicum L.	39	37
Bean, Pinto var. Phaseolus vulgaris L.[a]	22	25
Tomato, Sutton var. Lycopersicum esculentum L.[b]	27	23

[a]Bioassayed with Erwinia amylovora.
[b]Streptomycin was used as 1000 ppm in both spray and dust formulations.

pathogens. The validity of such an argument cannot be contradicted much as it applies to those diseases capable of establishing a foothold on almost any point on the plant. However, in some diseases, such as fireblight, for example, the necessary requirement for successful control of the disease under conditions obtainable in most localities in the country is to block the pathogen (Erwinia amylovora) in certain limited portals of entry such as the flowers. This can be accomplished by directing the toxicant into the portals of entry by suitable sprays or dusts.

Bordeaux sprays, fixed copper sprays, and monohydrate copper sulfate dusts are extensively used to control blight in California (3, 7). The attempts to determine the value of streptomycin dust in plant disease control were against fireblight of pear (1). Results obtained with the first formulations of streptomycin dusts were variable because of the use of bentonite and talc as carriers. Later it was demonstrated that these two types of carriers were immobilizing the streptomycin by irreversible adsorption, thus preventing its release from the carriers when it was placed on the surface of the leaf (2, 4). This difficulty was overcome when a pyrophyllite type of carrier, namely Pyrax ABB or Nuclay[3] was used instead of bentonite or talc.

It is well to bear in mind that the efficiency of dust formulations depend, in part, on the dampness of the surfaces to which they are applied and on the humidity of the air after the dust is applied. These conditions are not difficult to obtain in certain areas and with certain crops. Natti (5), working on control of downy mildew of broccoli with antibiotics, made an observation that the effectiveness of streptomycin was enhanced by exposing the sprayed plants to a constant misting. Dust formulations of streptomycin were tested by other workers on lima beans, etc. (8) and their performance was comparable to wettable formulations.

The pyrophyllite type of carrier does not inactivate streptomycin, but releases it gradually and over a longer period of time than do other agricultural carriers, such as gypsum. Gypsum releases streptomycin more rapidly, but over a shorter period of time (Tables 1 and 2).

The streptomycin released is capable of going into the plant cells almost immediately,

[3]Pyrax ABB -- Pyrophyllite produced by R. T. Vanderbilt Co., New York. Nuclay -- Pyrophyllite produced by the Kennedy Minerals Company, Inc., Los Angeles, California.

since it can be demonstrated in cell sap within 5 minutes after application. In the cell sap it constantly increases in concentration up to 24 hours, after which time the level gradually declines (Tables 3 and 4). Streptomycin absorption is not affected adversely by changes in temperature. The experiments on cucumber plants dusted and exposed to four different temperatures showed that even at such low temperatures as 2° to 5°C streptomycin is absorbed, at times even in a larger quantity than absorption at higher temperatures (Table 5). This appears to be in agreement with the conclusions of Schrödter (6), namely, that streptomycin treatments are considerably more effective at temperatures around 10°C than they are at temperatures exceeding 20°C.

Since streptomycin derived either from the dust or spray is absorbed by woody plants as well as by herbaceous types (Table 7) it can be used to control plant diseases affecting many different types of plants. Considering the fact that dust formulations of streptomycin are more economical than sprays, and with proper equipment are more easily applied, dusts merit wider use in plant disease control work. Also, the amount of available active antibiotic is not less but, rather, in some instances is even greater than in the wettable type.

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STABILITY OF STREPTOMYCIN DUST FORMULATIONS

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Abstract

Streptomycin dust formulations made in Pyrax ABB, Nuclay, sulfur, or lime rock did not appreciably diminish in potency during the 3 years of dry storage in paper bags at 23° to 25°C.

Streptomycin in dust form has proved to be effective against bacterial diseases and certain downy mildew diseases of plants. Growers in both California and Oregon use large quantities of streptomycin dust against fireblight of pear and walnut blight. The increase in the last few years of the annual consumption of this type of streptomycin formulation is based on its efficacy in disease control as well as on its definite economy, as compared with the highly expensive wettable formulations.

Since bentonite, frinite, and talc proved unacceptable as carriers for streptomycin (2), pyrophyllite is the chief ingredient suitable for streptomycin dusts at the present time (1). Calcium oxide, hydrated lime, lime stone, gypsum, and sulfur also can be used in certain special situations since they are quite compatible with streptomycin and do not interfere with absorption of the antibiotic from the formulations where applied on moist leaf surfaces of the plants (2).

Table 1. Release of streptomycin by streptomycin dust formulations stored for various periods of time at 23° to 24°C. Test organism: Erwinia amylovora. Inhibition zone: mm-dia. Last determination made on April 4, 1958.

Date of formulation	Formulator	Carrier	Diameter of inhibition zone around 13 mm paper disc: At start	At last determination
Feb., 1955	Merck, Sharp and Dohme	Pyrax ABB	25.4	25.6
Mar., 1955	Pacific Guano Co.	Sulfur	26.9	26.1
Mar., 1955	Pacific Guano Co.	Nuclay	23.4	22.7
Mar., 1955	Pacific Guano Co.	Nuclay	23.1	22.6
Mar., 1955	Pacific Guano Co.	Sulfur	25.8	24.7
Mar., 1956	Pacific Guano Co.	Nuclay	25.0	23.9
Mar., 1956	Pacific Guano Co.	Nuclay	22.8	20.3
Mar., 1955	Am. Cyanamid Co.	Pyrax ABB	27.4	27.3
Mar., 1955	Niagara Chemicals	Nuclay	23.1	22.0
Mar., 1955	Niagara Chemicals	Gypsum	23.4	21.2
Mar., 1955	Calif. Spray Chemicals	Nuclay	23.2	21.9
Mar., 1956	Calif. Spray Chemicals	Nuclay	25.7	25.4
Mar., 1957	Stauffer Chemical Co.	Nuclay	25.3	24.8
Mar., 1957	Stauffer Chemical Co.	Nuclay & Tricalcium Phosphate	27.0	26.4
Jan., 1955	Univ. of California	Nuclay	23.1	22.5
June, 1956	Univ. of California	Nuclay	23.4	23.2

To test the stability of streptomycin in different dust carriers, samples of the prepared streptomycin dusts were bioassayed and then stored in paper bags in a laboratory at room temperatures that fluctuated from 23° to 25°C. Both laboratory and commercial preparations were included in the tests. Determination of the strength of streptomycin was made by a bioassaying technique described elsewhere, which is based on the diameter of the clear zone around an impregnated filter paper disc (13 mm in diameter) placed on a seeded agar plate (2).

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Apparently the activity of streptomycin did not diminish in pyrophyllite, lime stone, sulfur, gypsum, and other carriers during the 3-year period of the test (Table 1). This longevity makes it possible to use the streptomycin dust formulations for more than one season.

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PRELIMINARY RESULTS FROM ANTIBIOTIC TREATMENTS OF PECAN NURSERY TREE ROOTS FOR CONTROL OF CROWN GALL (AGROBACTERIUM TUMEFACIENS)

John R. Cole[1]

Crown gall often causes injury to pecan trees. It was formerly considered to affect only nursery trees; but in recent years it has been found well established in orchards, especially on old trees which it may affect so severely as to kill them. On trees of bearing age the disease occurs mostly on the large roots and at base of the trunk, but occasionally it affects smaller roots also. Wartlike growths from a few inches to a foot or more in diameter, often extending several inches above the soil surface, characterize the development of crown gall on orchard trees. Because of their fragility, the galls are often broken off. The galls may then become scattered on top of the soil when the orchard is being cultivated, thus spreading the causal organism.

Control measures include the destruction of infected nursery trees, the removal of galls, and painting of the wounds with a mixture of one part of creosote to three parts of coal tar to prevent the spread of the organism to healthy parts of orchard trees.

In 1958, 45 nursery trees, the roots of which were moderately infected with crown gall, averaging two galls each and about 1 inch in diameter, were treated on March 7 with four antibiotics as follows: A, none (untreated checks); B, Terramycin; C, Agri-Strep; D, ACCO; E, Agri-mycin 100. The trees in each treatment were paired as to the amount of infection and their roots were soaked for 1 hour in water containing 400 ppm of the antibiotic material. Each treatment plot consisted of a single tree, which was replicated nine times. The trees were planted immediately after treatment on the Station grounds. Dry soil was placed in the holes around the roots and the trees were not watered until 24 hours after planting. Trees were cultivated to prevent weed growth and were irrigated when needed.

All 45 of the trees foliated, but some were later than others, regardless of the treatment. The foliage appeared normal on all trees and at no time did foliage or trees show signs of injury.

An examination of the trees above the ground on September 12, 1958 indicated that Terramycin had benefited the trees, since seven in treatment A; five in C; five in D; two in E were dead, while all nine in treatment B were green and apparently healthy.

On October 1 all 45 trees were dug and their roots carefully examined. All nine trees in treatment A had one or two galls and four in B had one or two small galls; one or two galls were also found on the roots in treatments C and D, as well as on eight trees in treatment E. Some trees in treatment A had two large galls. Bacteria of the causal organism were isolated from all trees in treatment A and two trees in treatment B. The trees in treatment B showed scars, where some galls had been destroyed.

One year's results of the dipping of pecan nursery tree roots in a solution of 400 ppm of the Terramycin antibiotic for 1 hour indicate that some control was obtained. Further tests are in progress.

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THE INCIDENCE OF THE RING SPOT VIRUS IN PEACH NURSERY AND ORCHARD TREES[1]

D. F. Millikan

Abstract

Indexing of nursery and orchard peach trees indicates that some trees are infected with the ring spot virus. The amount of infection was found to vary from about 8 percent in one orchard to over 20 percent in the case of a newly established scion block. Random sampling of unbudded Lovell seedling understocks showed about 15 percent infection, suggesting that these are a major source of inoculum. The vigor of the trees found to be infected does not seem to be visibly impaired and there appears to be no difficulty in locating disease-free clones of any one variety.

The ring spot virus was first described on peach by Cochran and Hutchins (2). However, aside from KenKnight's report (3), the disease on this crop has received little attention. Nonetheless, the similarity of ring spot isolates collected from cherry and peach (6) suggested a need for information concerning the incidence of infection in peach.

Both nursery and orchard sources were examined for the incidence of disease. Nursery sources were tested in order to evaluate the diseased conditions within Missouri nursery stocks. An orchard source was tested in an effort to determine the disease rating of exotic stocks. Once it became apparent that some disease was present in Missouri-grown nursery stock, then seedling stocks were also indexed.

METHODS OF INDEXING

Modifications of the Wisconsin and Oregon systems of indexing were used in the study. The former, described by Moore and Keitt (5), uses the sour cherry varieties Montmorency and English Morello as indicator hosts for sour cherry yellows and necrotic ring spot. The latter, described by Milbrath and Zeller (4), uses the flowering cherry varieties Kwanzan and Shirofugen as indicator plants. Only Montmorency and Shirofugen were used in the tests reported herein.

PRELIMINARY SURVEY IN 1952

Samples from two nurseries were collected and indexed on Montmorency. These samples included single trees of every variety listed by the respective nurseries. Results are listed in Table 1.

Table 1. Survey of incidence of ring spot infection in peach in two Missouri nurseries, 1952.

Nursery	Total number of trees	Number of healthy trees	Number of diseased trees
A	18	15	3
B	11	11	0

The incidence of ring spot infection found in 1952 varied from none in the case of nursery B to about 13 [16.7] percent in the case of nursery A. Although the number of trees tested was small, the results indicated that some disease was present in some locally grown nursery trees. No additional work was done until 1955.

[1]Journal Series Paper No. 1936, approved by the Director of the Missouri Agricultural Experiment Station.

INDEXING WORK SINCE 1955

In 1955 a more extensive survey for ring spot infection was initiated. Much of the renewed interest resulted from the establishment of a scion block by nursery C. Locally grown trees and trees purchased from an eastern nursery were included in this block. The results of this indexing are listed in Table 2.

Table 2. Incidence of the ring spot virus in the scion block of nursery C.

Variety	Number of trees	Number of trees infected				Number of healthy trees
		Mont. 1955	Shiro. 1956	Mont. 1956	Shiro. 1957	
Champion	3	0	0	0	0	3
Chinese Cling	2	0	0	0	0	2
Dixi Gem	5	0	0	0	0	5
Elberta	7	2	0	0	0	5
Early Elberta	5	0	0	0	0	5
Early Red Bird	2	0	0	0	0	2
Gage Elberta	5	2	0	0	0	3
Georgia Belle	4	-	2	0	0	2
Golden Jubilee	5	4	1	0	0	0
Halehaven	7	4	2	0	1	0
Indian Blood	2	1	0	0	0	1
J. H. Hale	6	2	0	0	0	4
July Elberta	7	0	0	0	0	7
Loring	5	2	0	0	0	3
New Day	5	0	0	0	0	5
Redhaven	6	1	0	0	0	5
Tulip	2	0	0	0	0	2
Totals	78	18	5	0	1	54

Twenty-four of the 78 trees tested were found to be infected with the ring spot virus. These were destroyed. Since Halehaven, Elberta and Golden Jubilee showed a high incidence of disease and are popular varieties, several nursery row selections were made and indexed on Shirofugen. Trees for this study were selected as every 10th tree in nursery rows of the desired variety. The results are listed in Table 3.

Table 3. Incidence of ring spot virus in nursery row selections of peach in nursery C, 1956.

Variety	Number of trees tested	Number of trees infected	Number of healthy trees
Elberta	6	0	6
Halehaven	5	1	4
Golden Jubilee	5	1	4

Results of indexing of the field selection of clones found to be carrying a high incidence of infection indicated that ring spot-free trees could be found with little difficulty. This also indicated that disease spread could be controlled from the scion source, but it did not eliminate the possibility of infection through seedlings as previously demonstrated by Cochran (1). Therefore, budsticks from 25 Lovell seedlings selected as every fifth tree from five short end rows were indexed on Shirofugen. About 2 months later four of these trees showed positive readings for ring spot infection. This suggested that seed transmission in this particular lot of seed might be as high as 16 percent.

In 1958 a long range study on virus spread was initiated in the University peach orchard. The orchard consists of 512 trees purchased from eight different sources located in seven

States. A total of 42 trees were found to be infected with ring spot, with no one variety completely infected.

DISCUSSION

Several years' study on the incidence of ring spot in peach nursery and orchard trees in Missouri indicated that some trees may be carrying the virus. Such trees are symptomless and if examined in the nursery row appear to be no less vigorous than adjoining healthy trees.

The percentage of infection found in the surveys ranged from none, in the case of a small sample from one nursery, to over 20 percent in a newly established scion block. Nursery row testing indicated that ring spot-free selections thought to be entirely infected could be found with little difficulty.

A random indexing of unbudded Lovell seedlings showed an incidence of infection amounting to about 16 percent. This indicates that any certification program must take into consideration seed as well as scion sources.

Indexing of an orchard planting of 512 trees purchased from eight different locations in seven States showed 42 trees or about 8 percent to be infected. No one variety was found to be completely infected, nor did there appear to be any particular correlation of infection with origin.

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KENTUCKY BLUEGRASS, POA PRATENSIS L., A NEW HOST OF THE BROMEGRASS MOSAIC VIRUS IN NATURE[1]

W. H. Sill, Jr., and Ren-jong Chiu[2]

The senior author for several years sought bromegrass mosaic virus, Marmor graminis McK. (4), in smooth bromegrass (Bromus inermis Leyss.) nurseries, planted small grain fields, and roadside grasses. The disease was found in Kansas in B. inermis only and was reported in this host from widely scattered areas (6). Although the virus has a rather wide possible host range (1, 3, 4), including dicotyledons as well as monocotyledons, it has never definitely been reported in nature from any species except B. inermis, although McKinney (1) may have found it in a barley hybrid.

In the summer of 1957 some Kentucky bluegrass plants, Poa pratensis L., showing mosaic symptoms and growing among clumps of virus infected smooth bromegrass plants were found in Morris County, Kansas. There was little stunting of the diseased bluegrass plants, but chlorotic mottling and striping were present on most leaves and occasional necrotic spots in the center of the chlorotic areas. Abrasive inoculations were first made to young Marquillo-Oro x Pawnee, Sel. No. 462666, wheat plants and to Golden Giant sweet corn seedlings. Symptoms, identical to those described for bromegrass mosaic virus developed on these hosts (1, 2, 5). Dicktoo barley and Golden Giant sweet corn seedlings were inoculated again with the same results. The characteristic top necrosis of sweet corn (2, 5), which is a diagnostic symptom for this virus, was particularly striking. Three other known collections of bromegrass mosaic virus from Kansas were then compared with the virus from bluegrass. These were all inoculated on the same day to the hosts mentioned above. Incubation periods and symptoms developing were essentially identical for all four collections and typical of bromegrass mosaic virus (1).

Although physical and chemical properties have not yet been compared, evidence thus far indicates that the virus from the naturally infected Kentucky bluegrass is bromegrass mosaic virus. Since there is no evidence of rapid spread of this virus by seed or insect vectors in the field, and no vectors are known (2), it is probable that the virus will survive and spread rather slowly in the perennial P. pratensis, much as it has in its other known natural perennial host, B. inermis (6). The present distribution and importance of the disease in Kentucky bluegrass is not known.

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THE INFLUENCE OF NITROGEN SOURCE AND CARBOHYDRATE CHANGE BY DEBUDDING AND GIRDLING ON BACTERIAL BLIGHT RESISTANCE CAUSED BY THE B_7 GENE IN COTTON

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Abstract

A strain of Upland cotton with the B_7 gene for bacterial blight resistance was used in experiments to study the influence of carbohydrate change and nitrogen source on resistance. Plants which were debudded became less tolerant while those with the main stem girdled became resistant. Eighty pounds of nitrogen per acre made tolerant plants become resistant. Nitrate nitrogen was more efficient in increasing resistance than was the ammonium form. The studies were made in the presence of Xanthomonas malvacearum races 1 and 2.

The use of natural plant resistance is the only known method for adequately controlling the bacterial disease of cotton which is caused by Xanthomonas malvacearum (E. F. Sm.) Dowson. For this reason a better understanding of the influence of environment on resistance is important for the plant breeder as well as for the grower.

Findlay (6) found that plant portions above the level at which the phloem had been removed from the stem by ringing were resistant to bacterial blight. In addition, he reported that plants from which flowers and squares were removed did not become more susceptible. The same writer reported experiments in which plants with waterlogged roots and plants sidedressed with quantities of ammonium sulfate became more susceptible. Bird (1, 2, 3) was able to duplicate Findlay's ringing experiment. However, he reported experiments in which resistance was increased and susceptible plants made resistant when sidedressed with quantities of ammonium nitrate.

MATERIALS AND METHODS

Cotton, strain E4-2-2-13, was grown on Miller clay loam soil located on the A. and M. Plantation in the Brazos Valley west of College Station. E4-2-2-13 carries the B_7 gene and is resistant to race 1 and tolerant to race 2 of X. malvacearum.

Nitrogen was applied as sodium nitrate, ammonium sulfate and alfalfa leaf meal in the 1957 test. The applications were made by sidedressing at the time of initial flowering at a rate to give 80 pounds of nitrogen per acre. When the nitrogen was applied the main plots were divided into subplots designated bud and debudded. Thus, the experimental design was randomized split plots. Six replications were used. The young squares were removed by hand from the plants in the debudded plots every 5 days.

Three weeks after nitrogen application the plants in the experiment were inoculated with inoculum composed of equal quantities of X. malvacearum races 1 and 2. Three weeks after inoculation the resulting infection was graded by using the system given by Bird (1, 2). In this system grades 1 to 4 represent degrees of resistance, 5 to 6 degrees of tolerance and 7 to 10 degrees of full susceptibility.

For the 1958 test the planting area was sidedressed with 200 pounds of ammonium nitrate 2 weeks before initiation of the experiment. The treatments made at the time of initial flowering were debudding, girdling of the main stem near the soil line, and the control. The experimental design was randomized blocks with eight replications. The plants were inoculated the day after girdling and debudding with inoculum composed of races 1 and 2. The resulting infection was graded 3 weeks after inoculation.

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RESULTS

The 1957 experiment was conducted to ascertain the influence of different nitrogen sources and carbohydrate increase by debudding on resistance. These results are given in Table 1. The plants with buds became resistant to both races when sidedressed with ammonium sulfate and sodium nitrate. Sodium nitrate was significantly more efficient than ammonium sulfate. Alfalfa leaf meal had no influence. The debudded plants, except those sidedressed with alfalfa leaf meal, became less tolerant. The change in resistance of the budded and debudded plants when sidedressed with sodium nitrate was about the same as for the control plants. The degree of change in the plants with ammonium sulfate was greater. This significant interaction is shown in Figure 1.

Table 1. The influence of nitrogen source and carbohydrate change by debudding on bacterial blight resistance.

Nitrogen source[a]	Average disease grade[b]		
	Buds	Debudded	Averages for nitrogen source
Control	4.35	4.81	4.58
Alfalfa	4.46	4.44	4.45
$(NH_4)_2SO_4$	3.72	4.57	4.14
$NaNO_3$	3.35	3.73	3.54
Averages for carbohydrate level	3.97	4.39	
L.S.D. for averages 1%		0.26	0.54
L.S.D. for averages 5%		0.19	0.39

[a] The nitrogen source-carbohydrate level interaction was significant at the 5% level.
[b] Based on a grading system ranging from 1 to 10, with 1 representing immunity and 10 full susceptibility.

The increased susceptibility of the debudded plants was not expected. For this reason the 1958 experiment was designed to compare carbohydrate increase in the leaves by debudding and by girdling the main stem. The results of this experiment are given in Table 2. The girdled plants became resistant, while the debudded plants became less tolerant.

DISCUSSION

Findlay (6) reasoned that a plant in the "carbohydrate or fruiting phase" would be more resistant to bacterial blight while one in the "nitrogen or vegetative phase" would be more susceptible. In the current experiments, as well as in those previously reported by Bird (1, 2, 3), the addition of excess quantities of nitrogen increased resistance, which is contrary to the results obtained by Findlay. The increase in susceptibility obtained by debudding agrees with Findlay's reasoning, since debudding maintained vegetative growth.

As shown by Eaton and Ergle (4) carbohydrates increase in the leaves of girdled plants while the nitrogen level remains the same. Findlay (6) and Bird (1, 2, 3) demonstrated that the above trend causes plants to become more resistant. Eaton and Joham (5) showed that in defruited plants the carbohydrate content of the roots increased almost three-fold, while little change was noted in the leaves. Their data also showed that at the same time the nitrogen content of the roots tended to increase while the trend, although not significant, was toward a lower nitrogen level in the leaves. Hall's (7) results showed that in leaves of debudded plants the carbohydrate level may increase slightly while the nitrogen level decreases. Bird (2) pointed out that with the carbohydrates remaining constant and with the nitrogen decreasing plants would become more susceptible. This would explain why debudded plants became more susceptible.

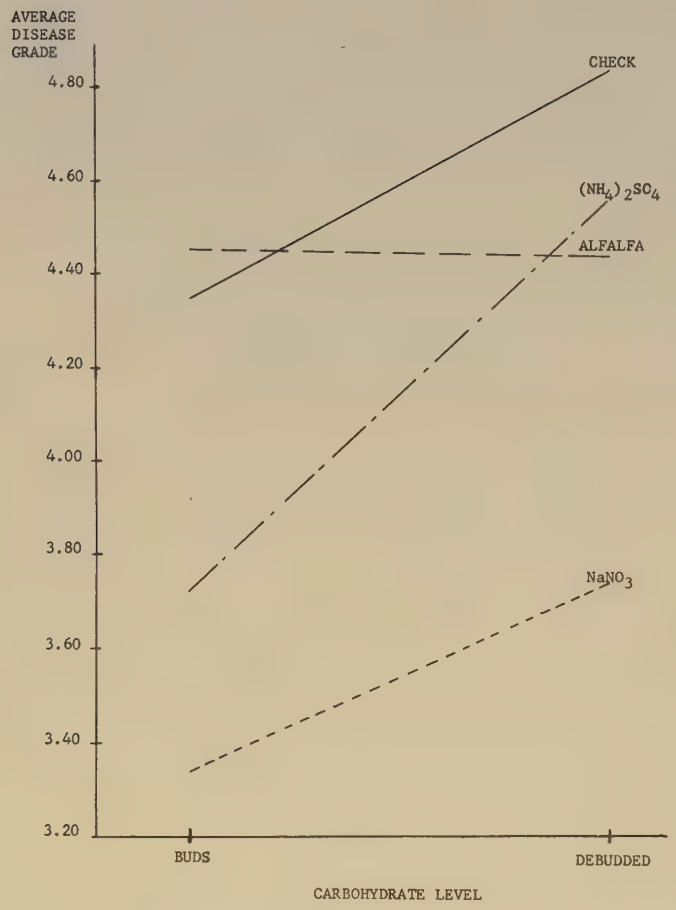

FIGURE 1. The differential response of bacterial blight resistance to nitrogen source and carbohydrate change caused by debudding.

Table 2. Comparison of carbohydrate change by girdling and by debudding.

Treatment		Average disease grade[a]
Girdled		2.19
Control		4.72
Debudded		5.25
L.S.D.	1%	0.5
	5%	0.4

[a] Based on a grading system ranging from 1 to 10, with 1 representing immunity and 10 full susceptibility.

The results reported here emphasize the importance of the relationship of nitrogen and carbohydrates in resistance to the bacterial blight disease of cotton. Genetic resistance and tolerance can be increased by providing plants with adequate nitrogen as indicated by a good green leaf color. These data point out that the nitrate form of nitrogen is more efficient than the ammonium form in improving resistance.

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COMPARISON OF FOUR SOIL FUNGICIDES IN THE GREENHOUSE FOR THE CONTROL OF SEEDLING DISEASES OF COTTON[1]

B. W. Kennedy and L. A. Brinkerhoff[2]

Summary

Zineb, PCNB (pentachloronitrobenzene), thiram, and captan when mixed with a steamed sandy loam each afforded relatively good protection against injury to cotton seed and seedlings by Rhizoctonia solani Kuehn provided the environment did not seriously retard germination and growth of the host.

Against Pythium debaryanum Hesse PCNB generally was not effective at rates as high as 1 part of active chemical to 5000 parts of soil (200 ppm). Captan, on the other hand, was effective against this fungus at 1 part of chemical to 40,000 parts of soil (25 ppm) when temperatures did not favor post-emergence injury.

Thiram was reasonably effective against both fungi when used at 1 part of chemical to 20,000 parts of soil (50 ppm). Zineb was also reasonably effective against both pathogens at 1 part of chemical to 5000 parts of soil (200 ppm) but was not effective at 1 part per 30,000 (33 ppm). PCNB was reasonably effective against R. solani at 200 ppm. The tests of the last material were not conclusive at lower rates.

Zineb, captan and PCNB retained their fungicidal effectiveness for 4 to 6 weeks in a non-steamed, moist sandy loam of pH 6.0. Control was somewhat less effective with thiram after 2 and 4 weeks.

None of the four fungicides was effective against either pathogen when low temperatures seriously retarded germination and growth of the host.

INTRODUCTION

Seedling diseases frequently cause considerable reduction in cotton stands in Oklahoma, as in other areas, when cotton is planted during cool or rainy weather. During the past 5 years a serious attempt has been made by a number of investigators to reduce seedling losses by applying fungicides with the covering soil at the time of planting (1, 4, 7, 8, 9, 12, 13, 15, 17). Although the results in general were promising, many failures were also recorded.

In artificially infested field tests Brinkerhoff et al. (4) obtained excellent control of R. solani with PCNB dust applied in the seed row. However, in several subsequent seed row tests in Oklahoma, PCNB was not effective against seed-rot and damping-off that occurred during cold spring rains. Thiram and zineb gave reasonably good protection under these conditions. Pythium spp. were repeatedly isolated during periods of cold rainy weather[3].

Cooper (5) obtained reasonably good control of pre-emergence injury to cotton in North Carolina with thiram and captan, but not with PCNB. More recently Whitehead and Brown (17) obtained somewhat similar results in artificially infested field tests in Missouri. Arndt (1) secured good protection against R. solani with PCNB in laboratory tests with sand cultures. Davis and Lund (6) found Pythium spp. to be the predominant pathogen isolated from diseased cotton seedlings in greenhouse tests with naturally infested soil at Stoneville, Mississippi. Neither PCNB nor zineb was effective at the rates used.

Fulton et al. (8) isolated R. solani and Fusarium spp. from early-planted cotton in Arkansas, whereas Glomerella gossypii Edg., Sclerotium bataticola Taub., and Rhizopus spp. were predominant in later plantings. PCNB apparently inhibited both R. solani and S. bataticola, but was not effective against the three other fungi. Ranney and Bird (14) reported that zineb,

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[3]Unpublished data.

captan and several other fungicides did not have the same effect at 70° as at 80° F in the greenhouse. The variable results with naturally infested soil were attributed to changes in the soil microflora. Isolations from infected cotton seedlings indicated that Fusarium spp. were predominant at 80°, whereas at 70° R. solani, Fusarium spp. and Pythium spp. were all active. In field tests (15) variable responses were obtained with several fungicides in soils of different pH levels. A mixture of captan, zineb and PCNB gave more uniform control than these fungicides used alone.

Sinclair (16) found that 17 different fungicides gave considerable variation in the control of post-emergence injury from R. solani in greenhouse tests in non-sterilized, artificially infested soil. PCNB gave as consistent results as any of the other chemicals.

In the present study an attempt was made to ascertain fungicidal and residual properties of zineb, thiram, captan, and PCNB in both steamed and non-steamed soil under greenhouse conditions. Chemicals were thoroughly mixed with soil and cotton seeds were planted after a lapse of time varying from 0 to 6 weeks. R. solani and P. debaryanum were used as test pathogens.

GENERAL METHODS AND MATERIALS

Soil for the fungicides tests was obtained from a fertile bottom land where cotton had been grown for a number of years. It was classified as Port sandy loam, and tests showed an organic matter content of 0.9 percent and pH of 6.0. Steam was used for partial sterilization.

The fungicides tested were wettable powders of the composition shown in Table 1. Each formulation was mixed first with fine dry sand and then thoroughly mixed with air-dried soil or in some instances with screened moist soil.

Table 1. Description of chemicals used in soil-fungicide tests.

Fungicide[a]	Active principle		Manufacturer
	Name	Percent	
Zineb	Zinc ethylene bisdithiocarbamate	65	Rohm & Haas
Thiram	Tetramethylthiuramdisulfide	75	DuPont
Captan	N-trichloromethylthiotetrahydrophthalimide	50	Calif. Spray
PCNB	Pentachloronitrobenzene	75	Olin Mathieson

[a]The zineb was Dithane D-78; thiram was Tersan 75; and captan was Orthocide 50.

The commercial formulations of wettable powders were first tested for phytotoxicity at four rates: 1 part to 20,000 parts of soil, 1:10,000, 1:5,000, and 1:2,000. The 1:20,000 rate was calculated to approximate field tests in which the chemicals had been applied as sprays at 5 pounds per acre in the seed furrow and covering soil at the time of planting. Rates for subsequent tests were based on the results of this phytotoxicity test.

A single lot of acid-delinted seed of the variety Deltapine 15 was used for all tests. A plot consisted of a single flat (15 x 18 inches) planted with 100 seeds or a 6-inch pot planted with 10 seeds. Treatments were randomized and replicated, and data taken so that it could be analyzed statistically.

Methods used to infest the soil were similar to those described for studies with R. solani (3). Both R. solani and P. debaryanum were grown for 2 weeks on steamed-sterilized grain sorghum seed and then mixed with the soil as whole grain, or hyphal suspensions were prepared in a blender and sprinkled over the surface of the soil at the seed level. Hyphal suspensions of each pathogen were prepared by homogenizing the contents of a 125-ml flask culture, straining through cheese cloth and adding sufficient water to make 1 liter. Fifty ml of this suspension was used for R. solani and 90 ml for P. debaryanum per flat, or 5 ml for each per 6-inch pot. Ten seeds of the whole grain culture were thoroughly mixed with the soil of each 6-inch pot when this method was used. Plantings were made immediately after infesting with the hyphal suspensions and 20 days after infesting with the whole grain.

RESULTS

Phytotoxicity Test

Wettable powder of each of the formulations supplied by the respective manufacturers was mixed with non-steamed soil at the rate of 1 part formulation to 20, 000 parts of soil, 1:10, 000, 1:5, 000 and 1:2, 000. The test was made in the greenhouse when summer temperatures ranged from 75° to 110° F.

With temperatures favoring rapid germination none of the chemicals seriously reduced the percentage of germination. However, growth was much retarded by the higher rates of thiram and considerable chlorosis was associated with captan (Figs. 1 and 2). As observed by Kortsen (10), captan accelerated seedling emergence by about 24 hours when temperatures were favorable for rapid germination.

FIGURE 1. Six-day-old cotton seedlings in soil in which thiram (75 percent) was thoroughly mixed prior to planting at the following rates: A. 1:20, 000; B. 1:10, 000; C. 1:5, 000; D. 1:2, 000. Note delayed emergence at the higher rates.

FIGURE 2. Cotton seedlings grown in soil in which captan (50 percent) was thoroughly mixed prior to planting at the rate of 1 part chemical to 20, 000 parts of soil. Note chlorosis on the margins of the cotyledons.

Test with Fungicides at Different Rates

In this test flats of steamed soil were artificially infested with P. debaryanum or R. solani after the chemicals had been mixed with the soil. Rates for thiram and captan were reduced since phytotoxicity had been encountered in the phytotoxicity test. The culture of R. solani was not virulent. Greenhouse temperatures which ranged from about 60° to 80° F were very favorable for the expression of the disease.

Captan gave relatively good protection, but the other materials almost none (Table 2).

Table 2. Cotton seedlings surviving 19 days in treated steamed soil infested with P. debaryanum.

Chemical name and rate (formulation: soil)	Seedlings surviving (percent of seed planted)	Chemical name and rate (formulation: soil)	Seedlings surviving (percent of seed planted)
Zineb:		Captan	
1:20,000	0	1:45,000	46
1:15,000	0	1:40,000	59
1: 7,000	0	1:30,000	65
1: 3,000	10	1:20,000	59
Thiram:		PCNB:	
1:40,000	1	1:20,000	0
1:30,000	0	1:15,000	0
1:25,000	0	1:10,000	0
1:20,000	0	1: 7,000	1

Additional Tests in Steamed Soil

Six-inch clay pots of steamed soil were inoculated with 10 seeds of a grain sorghum culture of either R. solani or P. debaryanum, after which all pots were placed in the greenhouse and kept moist for 20 days. The fungicides were then thoroughly mixed with the soil, the soil repotted and seeds planted. Temperatures ranged between approximately 70° and 85° F, which favored relatively rapid germination of the seed.

The results are presented in Tables 3 and 4.

Table 3. Relative effectiveness of four fungicides as protectants against injury caused by P. debaryanum in steamed soil.

Treatment	Rate[a] (Chemical: soil)	Cotton seedlings as percent of seed planted		
		Emerged after 12 days	Surviving after 18 days	Free of lesions after 22 days
Infested soil:				
Zineb	1: 5,000	83.3	80.0	60.0
Thiram	1:20,000	70.0	70.0	26.6
Captan	1:60,000	50.0	36.6	0.0
PCNB	1: 5,000	0.0	0.0	0.0
No chemical		0.0	0.0	0.0
Non-infested soil:				
No chemical		86.6	86.6	86.6

[a]Calculated on basis of active ingredient of the chemical.

An analysis of variance showed a high level of significance for treatments for emerged and surviving seedlings. PCNB gave no protection against P. debaryanum but was reasonably effective against R. solani. Captan at the rate used was not effective against post-emergence damage by either pathogen.

Residual Tests

Two tests were made in flats with each of the four chemicals mixed with moist non-steamed soil and held for 0, 1, 2, and 4 weeks before infesting and planting. From the time of treating until 4 days after infesting, the treated soil was kept moist and held at 65° to 70° F. Post-emergence temperatures ranged from approximately 80° to 110° for the first test, which was made in the summer and from 60° to 80° for the second test. In a third test in 6-inch pots

Table 4. Relative effectiveness of four fungicides as protectants against injury caused by R. solani in steamed soil.

Treatment	Rate[a] (Chemical: soil)	Cotton seedlings as percent of seed planted		
		Emerged after 12 days	Surviving after 18 days	Free of lesions after 22 days
Infested soil:				
Zineb	1: 5,000	76.6	76.6	60.0
Thiram	1:20,000	90.0	86.6	76.6
Captan	1:60,000	90.0	20.0	0.0
PCNB	1: 5,000	73.3	73.3	70.0
No chemical		20.0	3.3	0.0
Non-infested soil:				
No chemical		80.0[b]	80.0[b]	53.3[b]

[a]Calculated on basis of active ingredient of the chemical.
[b]One replicate had infected seedlings indicating that contamination had occurred at some time during the test.

the chemicals were mixed with air dried soil placed in the greenhouse with the moisture level maintained as for growing plants for 0, 3, and 6 weeks before infesting and planting. Temperatures for the last test including the period when the seed germinated ranged between 70° to 85°.

Data for the test made during the summer are presented in Table 5. R. solani again was not pathogenic apparently because a non-virulent isolate was used. Against P. debaryanum, zineb (65 percent) and PCNB (75 percent) at 1:20,000 were relatively ineffective regardless of the length of time the soil was treated. Captan (50 percent) afforded good protection and was as effective after 1, 2, and 4 weeks as immediately after treating. Thiram (75 percent) also gave good protection but was somewhat less effective after being in the soil 2 and 4 weeks.

Table 5. Relative effectiveness of four fungicides mixed with non-steamed soil and tested against P. debaryanum after 0, 1, 2, and 4 weeks.

Treatment	Rate[a] (Chemical: soil)	Surviving cotton seedlings as percent of seed planted (Time chemicals in soil before infesting and seeding: in weeks)				
		0	1	2	4	Mean
Infested soil:						
Zineb, 65%, formulation	1:20,000	5.0	8.5	1.5	8.0	5.8
Thiram, 75%, formulation	1:20,000	87.0	78.5	60.5	45.5	67.9
Captan, 50%, formulation	1:20,000	76.5	83.0	81.5	72.5	78.4
PCNB, 75%, formulation	1:20,000	31.0	30.5	7.0	16.0	21.1
No chemical	1:20,000	3.0	8.0	3.0	5.5	4.9
Non-infested soil:						
No chemical		88.5	87.5	77.0	90.5	85.9

[a]Rates calculated on the basis of 5 pounds per acre of each commercial formulation for 40-inch row spaces.

In a second test identical to the above except for environmental conditions (shorter day length at temperatures of 60° to 75°) severe post-emergence injury by the pathogen was evident. In flats treated with captan a mean of 42.4 percent cotton seedlings survived, with zineb 13.8, with thiram 9.9, with PCNB 16.1; on the infested check 0.5 and on the naturally infested check 35.5 percent survived. Captan was as effective after 4 weeks as immediately after treating the soil.

Data from the third test are presented in Table 6. Again temperatures retarded emer-

gence of cotton and favored severe injury from the pathogens. Control in the artificially infested soil was relatively poor. An analysis of variance showed no statistical significance for the time the chemicals were in the soil.

Table 6. Comparative effectiveness of four fungicides mixed with nonsteamed (air-dried) soil held for various periods before infesting with R. solani or P. debaryanum.

Description of soil	Rate[a] (chemical: soil)	Survival after 18 days (percent of seed planted) in soil held indicated period before infesting and seeding (in weeks)			
		0	3	6	Mean
Soil infested with R. solani after treatment and holding:					
Zineb	1: 5,000	26.6	26.6	30.0	27.7
Thiram	1:20,000	33.3	13.3	12.0	18.8
Captan	1:60,000	13.3	26.6	13.3	17.7
PCNB	1: 5,000	23.3	50.0	46.6	40.0
No chemical		3.3	3.3	6.6	4.4
Soil infested with P. debaryanum after treatment and holding:					
Zineb	1: 5,000	23.3	3.3	16.6	14.4
Thiram	1:20,000	6.6	0.0	10.0	5.5
Captan	1:60,000	6.6	3.3	16.6	8.8
PCNB	1: 5,000	3.3	3.3	16.6	7.7
No chemical		0.0	26.6	0.0	8.8
Naturally infested soil treated and held:					
Zineb	1: 5,000	96.6	100.0	100.0	98.8
Thiram	1:20,000	90.0	93.3	83.3	91.1
Captan	1:60,000	43.3	83.3	93.3	76.6
PCNB	1: 5,000	60.0	66.6	83.3	70.0
No chemical		16.6	46.6	60.0	41.1

[a]Calculated on basis of active ingredient of the chemical.

DISCUSSION

The results obtained in steamed soil in the greenhouse confirmed field observations that PCNB generally was effective against R. solani but not against P. debaryanum. Partial control was obtained in non-steamed soil against P. debaryanum in one test held at relatively high temperatures during the post-emergence period.

Captan and thiram appeared to be effective against both P. debaryanum and R. solani at considerably lower rates than zineb provided that growth of the host was not seriously retarded by low temperatures. At relatively high temperatures (75° to 110° F) captan and thiram exhibited a marked degree of phytotoxicity to germinating cotton when used at rates exceeding 1 part active chemical per 20,000 parts of soil.

In the slightly acid non-steamed moist soil used in these studies decomposition of the fungicides over a 4 to 6 weeks period did not appear to be of any significance except possibly for thiram. The frequent failures in the field of these materials to control post-emergence injury would not appear to be due to chemical decomposition of the fungicides.

There were striking differences in disease control in artificially infested sterilized soil as well as in non-sterilized soil for each of the four chemicals under different environmental conditions. At relatively low temperatures (60° to 75° F) disease control was frequently inadequate. Leach (11) reported that watermelon, a high-temperature crop like cotton, escaped

infection by either P. ultimum Trow or R. solani at 95° but was severely infected as the temperature was lowered.

It would appear that many of the same factors responsible for the success or failure of seed protectants of cotton also operate for the success or failure of these materials when they are thoroughly mixed with the soil. When the environment was not favorable for growth of cotton the protective value of each of the chemicals was reduced. Control was marked at temperatures that favored relatively rapid growth of the host. It is suggested that these same host-pathogen relationships may explain many of the inconsistencies observed in the field when fungicides are applied with the covering soil at the time of planting.

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YELLOW STRAPLEAF DISEASE OF CHRYSANTHEMUM[1]

C. R. Jackson and S. S. Woltz[2]

In recent years a new disease has appeared sporadically in Florida chrysanthemum plantings. The disease, which for the past 5 years affected plants in only a few growers' plantings, has increased considerably during the summer and early fall of 1958. In a recent survey of the major pompon chrysanthemum-growing areas of Florida the disease was found in almost every large commercial planting. Less than 10 percent of the plants in any single planting were affected, and in many cases fewer than 0.1 percent diseased plants were noted. The cause of the disease is not known. The name yellow strapleaf is suggested because pale yellow, narrow leaves are a conspicuous symptom of the disease. The purpose of this report is to outline the symptoms of the disease and cultural practices which may be important in this problem.

SYMPTOMS

The disease appears most commonly after rooted cuttings have become established in beds, usually about 26 days after planting, so that the affected plants have 7 to 10 apparently normal basal leaves. Initial symptoms are seen in young leaves during the first days of their expansion and separation from the axillary bud following removal of the terminal growing point. These leaves fail to expand normally and become involuted with incurved tips. Frequently if the "pinch" is delayed or omitted, the symptoms will develop from the terminal growing point. As these narrow, pale yellow leaves slowly expand they become less involuted and assume a dull yellow or ivory color. At this stage leaves are frequently narrow, very elongated, slightly brittle, with small lobes, serrations, or entire margins (Fig. 1).

A characteristic symptom is the severe retardation of the axillary buds following the removal of the terminal growing point (Fig. 2). The axillary buds become yellow and slightly swollen (Fig. 3); new leaves diverge very slowly and internodes are short. The upper portions of the stems of affected plants are frequently slightly larger in diameter and the entire stem becomes abnormally hard and brittle.

Only slight variations in symptoms occur among the varieties commonly cultivated. Greatest differences are found in the degree of stunting in plants of the same age and in the shape of leaves. The latter difference is due, in part, to varietal differences in size and shape of leaves. The disease occasionally occurs in older plantings that are approaching flowering, even after buds are visible.

Diseased plants may remain stunted and yellow for 6 to 8 weeks and during this period may grow only 4 to 6 inches. Leaves occasionally become necrotic at the tips and along the margins. Necrotic flecks may develop on the tips of axillary buds. Root development is normal and in some instances appears to be slightly greater in diseased plants. Stunted plants often resume an almost normal growth rate after varying periods of time. Renewed growth occurs mainly by the elongation of several axillary buds. The existing strap-like leaves become larger but never assume normal shapes. Chlorotic tissues become green and new growth is normal in most respects but recovered plants are seldom as tall as healthy plants at flowering time. Flower heads produced by plants which have resumed growth are frequently small and distorted with few expanded ray petals. The centers of heads are abnormally compact and green, resulting partially from the abnormal development of involucral bracts around disc flowers.

DISTRIBUTION IN PLANTINGS

Pompon chrysanthemums are cultivated in beds, generally 80-125 x 3 1/2-4 feet in size, which support 1300 to 1500 plants. The occurrence of the disease in successively planted beds is irregular and no consistent patterns are found within beds. The disease may affect all plants in 15 to 20 feet of a bed, while plants of the same variety and age in adjacent beds may be unaffected. Or the plants may show a definite height gradation due to the presence of diseased plants; for example, sloping from the apparently normal center plants to stunted

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FIGURE 1. Leaves of Iceberg variety from upper portions of stems; top -- healthy, middle -- diseased, bottom -- from recovering plant.

FIGURE 2. Iceberg variety, 10 weeks old; left -- healthy, right -- stunted plant with strap-like upper leaves.

FIGURE 3. Axillary buds from plants shown in Figure 2; left -- healthy, right -- diseased.

plants along both edges, grading from normal marginal plants along each edge to severely stunted plants in the center of a bed, or grading from normal plants along one edge to stunted plants along the opposite edge. Such conditions may occur along the entire length of a bed. Two or three intermediate size plants separate the stunted and normal plants in these cases. In addition, a single diseased plant occasionally is found in a bed of normal, vigorous plants or a single healthy plant may be found in a bed of diseased plants.

The two pompon varieties most commonly and severely affected are Blue Chip and Iceberg. Jetfire variety, at the other extreme, shows considerable resistance.

TEMPERATURE AND SOIL FACTORS

During the summer and early fall when high temperatures prevail, the incidence of yellow strapleaf is higher than during the rest of the year. Recovery of stunted plants and a decrease in the incidence of the disease appear to be associated with the advent of lower temperatures in the fall, but this relationship has not been demonstrated experimentally.

The disease develops in plants growing on various soil types throughout the State. These soils are all sandy and relatively porous and are usually amended by the addition of peat. The soils are, with few exceptions, treated with chemical fumigants or steam before planting. No relation between the method of soil treatment and the incidence of disease has been found.

Plants used in pot cultures have not shown symptoms of the disease. Severely stunted plants recover rapidly when transplanted from a bed, potted in soil taken from around diseased plants, and placed in the greenhouse. Recovery is much slower, however, if the potted plants are placed on the surface of the bed where the disease is present.

Treatment with known nutrient elements has not resulted in recovery or marked improvement regardless of whether the element was applied as a foliar spray or as a soil drench.

The possibility of the inadvertent application or presence of herbicides has been investigated without indication that such may be the cause. Attempts have been made by the writers and other workers in Florida to transmit a causal organism by grafting diseased scions onto healthy stocks of the same or different variety. The disease has never been produced in this way and new growth from affected scions is normal. Most tip cuttings from diseased plants develop into normal, vigorous plants although root growth is slower initially, compared with unaffected cuttings.

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SYMPTOMS OF MINERAL DEFICIENCY IN THE LOWBUSH BLUEBERRY[1]

C. L. Lockhart[2]

Abstract

Lowbush blueberries were grown in sand culture with mineral solutions deficient in the elements N, P, K, Mg, Ca, S and Fe to determine deficiency symptoms. Symptoms resulting from deficiency of the major elements were found to be similar to those that were due to similar deficiency in other crops.

Necrotic spots associated with symptoms of deficiency of Mg and P or the flecking accompanying Ca and N deficiencies were not considered to be similar to the leaf spotting of unknown origin occurring on lowbush blueberry plants under field conditions.

The growth of blueberry plants calculated on a dry weight basis was significantly less in plants grown in solutions deficient in Ca, N, Mg, S and P than that of plants grown in the complete nutrient solution. There were no significant differences in growth between K- and Fe-deficient plants and the control plants.

INTRODUCTION

Considerable leaf spotting of undetermined origin appears on the leaves of lowbush blueberry plants just before ripening of the fruit. A study of the plants' mineral requirements was undertaken to determine whether a deficiency of any of the major elements was the cause of this leaf spotting.

MATERIALS AND METHODS

Rhizome cuttings from a single blueberry clone of Vaccinium angustifolium var. laevifolium House were grown in greenhouse soil until shoots were about 1 inch high. The plants were then removed, washed in distilled water, and transferred to sand cultures. The sand cultures consisted of quartz sand (99.7 percent silicon oxide, 20-30 mesh) in 7-inch clay pots lined with polyethylene sleeves. An inverted watch glass was placed in the bottom of each pot to prevent loss of sand through the drainage hole.

Full nutrient solutions and those deficient in N, P, K, Mg, Ca, S and Fe were prepared in glass distilled water. The concentration and amount of each chemical used are listed in Table 1 and the compositions of the nutrient solutions are given in Table 2. The full nutrient solution was used for the control. The solutions were applied by the continuous-flow method of Shive and Stahl (2). This supplied each plant with 400 to 500 ml of solution per day.

The pH of nutrient solutions except for the full nutrient and Fe-deficient solutions ranged from 4.0 to 4.9 and were left unadjusted. The pH of full nutrient and Fe-deficient solutions were 6.8 and 5.5, respectively, and were adjusted to pH 4.0 with HCl.

The treatments, replicated four times in randomized blocks, were applied for 23 weeks. The sand cultures were flushed with distilled water once a week to remove excess salts.

Measurements of shoot length were made at weekly intervals and observations were made on deficiency symptoms as they appeared. At the end of the experiment the tops and roots were removed from the rhizomes and their weights determined after they were dried in an oven at 50°C for 40 hours.

GROWTH OF BLUEBERRIES IN SOLUTIONS DEFICIENT IN NUTRIENTS

The data presented in Figure 1 show that the growth of blueberries, as indicated by shoot length, was retarded mostly in plants that had been given solutions deficient in Ca, and that growth improved progressively in solutions deficient in N, S, P, Mg, K, in the control and in

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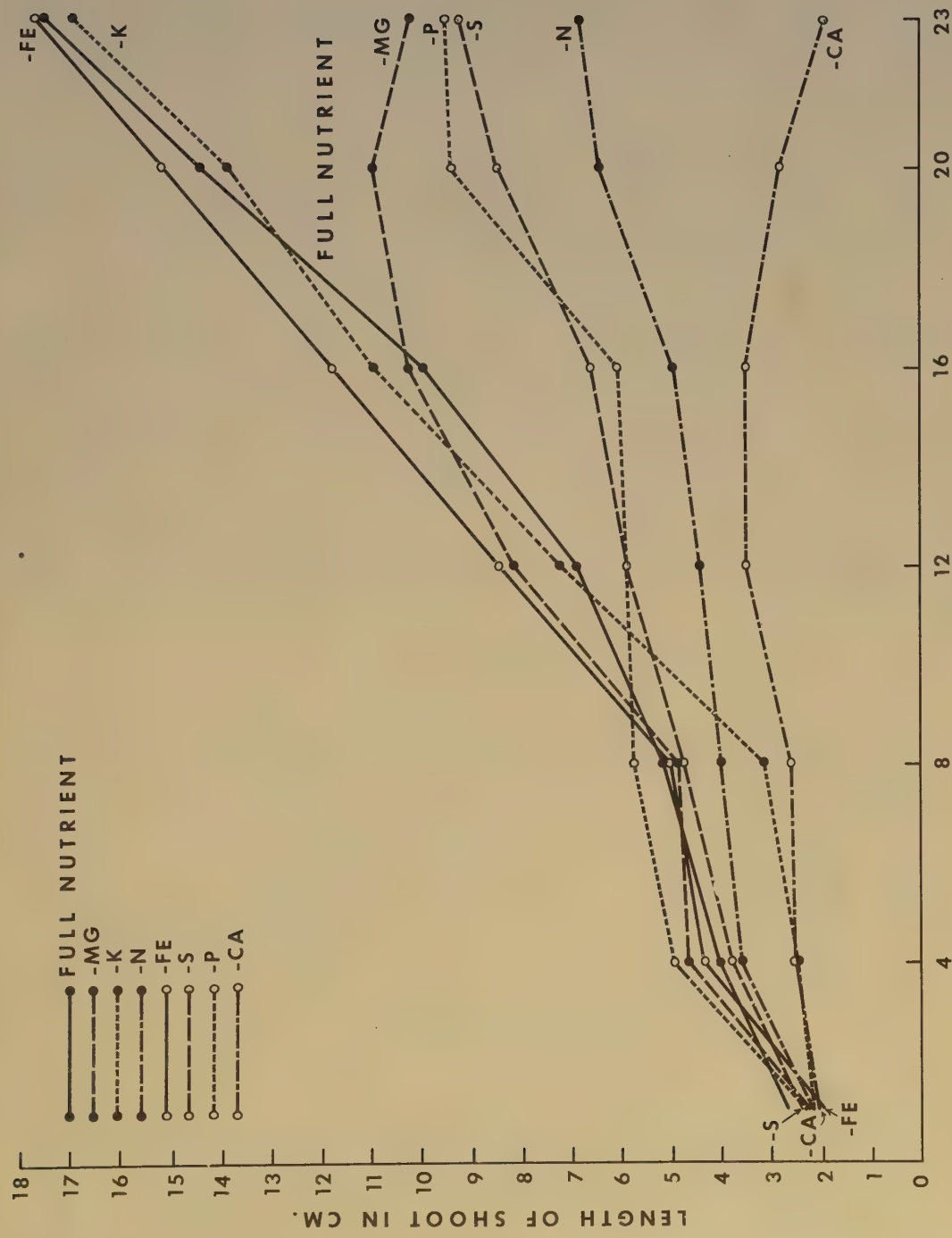

FIGURE 1. Growth of blueberries in solutions deficient in nutrients.

Fe-deficient solution. Blueberry plants receiving solutions deficient in S and P developed a branched type of growth, and consequently weighed more (Table 3) than those that received an Mg-deficient solution.

The data presented in Table 3 show that the dry weights of blueberry plants that received solutions deficient in Ca, N, Mg, S and P were significantly less than the weight of the control. There were no significant differences between the K- and Fe-deficient blueberry plants and control.

Table 1. The concentration and amount of chemical salts used in nutrient solutions.

Symbol	Compound	Molar concentration	Ml/liter used (see Table 2)
A	$Ca(NO_3)_2 . 4H_2O$	1	5
B	KNO_3	1	5
C	$MgSO_4 . 7H_2O$	1	2
D	KH_2PO_4	1	1
E	$CaH_4(PO_4)_2 . H_2O$	0.01	50
F	K_2SO_4	0.5	20
G	$CaSO_4 . 2H_2O$	0.01	200
H	$Mg(NO_3)_2 . 6H_2O$	1	2
I	Iron tartrate	0.002	1
J	Trace element solution[a]	0.025	1

[a] $MnCl_2 . 4H_2O$, 1.81 gm; H_3BO_3, 2.86 gm; $ZnSO_4 . 7H_2O$, 0.22 gm; $CuSO_4 . 5H_2O$, 0.08 gm; $H_2MoO_4 . H_2O$, 0.09 gm; per liter.

Table 2. Compositions of nutrient solutions.

Nutrient solution	Contents (see Table 1)
Full nutrient	A, B, C, D, I and J.
Minus nitrogen	C, E, F, G, I and J.
Minus phosphorus	A, C, F, I and J.
Minus potassium	A, C, E, I and J.
Minus calcium	B, C, D, I and J.
Minus magnesium	A, B, D, F, I and J.
Minus sulfur	A, B, D, H, I and J.
Minus iron	A, B, C, D and J.

DEFICIENCY SYMPTOMS

The foliage of the nitrogen-deficient plants was a paler green than normal and showed an interveinal flecking mainly along the margins of the leaves. This symptom was followed by a defoliation of the lower leaves and stunting of the entire plant.

The first evidence of phosphorus deficiency was a slight interveinal chlorosis with a dark-green background around the veins. In advanced stages the younger blueberry leaves became pink to reddish in blotches progressing inwards from the leaf margins. Necrotic spots appeared on the older terminal leaves. The spots coalesced and the leaves died within a week after the spots first appeared. Dark blotches sometimes appeared on the foliage and the plants took on a bushy appearance.

Symptoms of potassium deficiency, which were slow in appearing, were chlorotic blotches, reddish flecking and red veins. Later the foliage became bluish green, and finally purplish interveinal blotches formed on the lower leaves. In the advanced stages the purple color progressed upward to affect the whole plant to a lesser degree. The plants showed good growth.

Magnesium deficiency caused interveinal red to brown blotches in the central portion of the middle and lower leaves. Sometimes necrotic spots developed in these blotches as the

Table 3. The dry weights of tops and roots of blueberry plants.

6 Solution	Average weight of replicates in grams		
	Tops	Roots	Total[a]
- Ca	0.09	0.05	0.14
- N	0.37	0.16	0.53
- Mg	0.53	0.18	0.71
- S	1.14	0.45	1.59
- P	1.27	0.34	1.62
- K	2.48	0.59	3.07
Control	3.34	1.20	4.54
- Fe	3.84	0.88	4.72

[a]L.S.D.: 5% level, 1.80; 1% level, 2.46.

symptoms appeared on the upper leaves. In the advanced stages defoliation started with the middle leaves and was followed by defoliation of the lower leaves. Sometimes a less pronounced reddening similar to that found on phosphorus-deficient plants occurred on the younger leaves.

The foliage of calcium-deficient plants was slightly chlorotic with red to dark flecks which turned to dark brown blotches and coalesced as the leaves curled up in the advanced stages and died. Growth of plants was very poor.

Sulfur deficiency appeared as a deep red to purple color on the younger blueberry foliage. This reddish color slowly faded leaving four or five of the terminal leaves affected. Slight interveinal chlorosis and dark flecking occurred occasionally. Blueberry plants showed a bushy type of growth.

The foliage of iron deficient plants was slightly chlorotic with red flecking in the younger leaves of some plants. The plants showed good growth.

DISCUSSION AND CONCLUSIONS

The necrotic spots which occurred on the older terminal leaves in the advanced stages of phosphorus deficiency appeared, at first, somewhat similar to leaf spotting of blueberries under field conditions. However, in the field the spots have never been seen to coalesce and cause the defoliation observed in these greenhouse experiments. Neither the flecking of the foliage which accompanied nitrogen and calcium deficiencies nor the necrotic spots on magnesium-deficient plants resembled the leaf spotting of blueberries in the field.

Symptoms accompanying deficiency of major elements in lowbush blueberry plants appear similar to those occurring on other crops, Bahrt et al. (1). Bahrt's experiments included citrus, apple and peach crops.

Growth of blueberry plants calculated on a dry weight basis was significantly less in those plants receiving solutions deficient in Ca, N, Mg, S and P than in those of the control. There were no significant differences in the growth of K- and Fe-deficient plants and that of the control. The excellent growth of blueberry plants receiving solutions containing little or no iron indicated the low iron requirements of the lowbush blueberry.

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GUMBOIL DISEASE OF APRICOT

H. E. Thomas and George Nyland

Summary

Gumboil is suggested as the name for an undescribed rough bark condition in apricot in California. The disease, found in 5 to 7 percent of the trees in some orchards of the Royal variety, shows as discrete swellings in the bark of younger limbs and extremely rough cracked bark of the trunk and older branches. Masses of brown gum occur in the wood and bark in necrotic pockets under the swellings. In older branches these gum pockets extend from the inner wood to the surface of the bark. Affected trees are stunted to about one-half normal size. All transmission tests, some of which were begun in 1939, have been negative. The condition is perpetuated in buds from diseased trees. The nature of the disease is not known but may be of genetic origin.

The name "gumboil" is suggested for a disease of apricot that is known to occur in several apricot districts of California -- in Contra Costa, San Benito, San Joaquin, Santa Clara, and Stanislaus Counties. Diseased trees have been found only in orchards of the Royal variety. In the orchards mapped incidence varied from 4 to 7 percent. All attempts to transmit the disease by grafting have failed. Transmission data are not considered complete, but a report of this viruslike disease may be of interest at this time.

SYMPTOMS

The disease is most striking in trees about 10 years old or older. Affected trees are smaller and less fully branched than unaffected trees. There is some die-back of small shoots, and terminal growth is usually less than normal.

The most conspicuous symptom is the extremely rough, cracked bark of the trunk and older branches (Fig. 1, A, B). This roughness becomes less pronounced toward the younger wood, where it gives way to more or less discrete swellings in the wood and bark. Removing surface bark from these areas reveals masses of brown gum in necrotic pockets (Fig. 1, C). In older branches these pockets extend from the inner wood to the surface of the bark (Fig. 1, D). On 3- to 6-year-old shoots, only discrete blisters, or gumboils, can be seen. Gumboils are difficult to find in wood younger than 3 years.

On myrobalan roots, affected trees are markedly overgrown at the union (Fig. 1, B). Such trees exhibit excessive development of root suckers. The understocks are much smaller than those of normal trees of the same age. Affected trees are usually the first in the orchard to show signs of water stress between irrigations.

Fruits are fewer on affected trees than on healthy trees, but otherwise appear normal for the Royal variety. Typically they mature a few days earlier than fruits on normal trees.

Distribution of diseased trees in the orchard suggested that propagation was the means by which the disease was spread. Most affected trees of the same age showed the same degree of symptom expression. A tree was occasionally found with symptoms more severe or advanced on one side or in one or two main branches than in the others. No partially affected trees were found. Affected trees occurred singly or in scattered groups of two or more. In one case it was possible to trace the origin of affected trees to a specific orchard containing diseased trees.

Transmission Tests

Transmission tests to determine whether gumboil was associated with a virus were begun in 1939, at Berkeley. Healthy scions of Royal and affected scions were grafted on myrobalan 29. No evidence of transmission was obtained in 19 years, but the disease was perpetuated in these trees and in a tree where affected scions were grafted on an apricot seedling. In 1953,

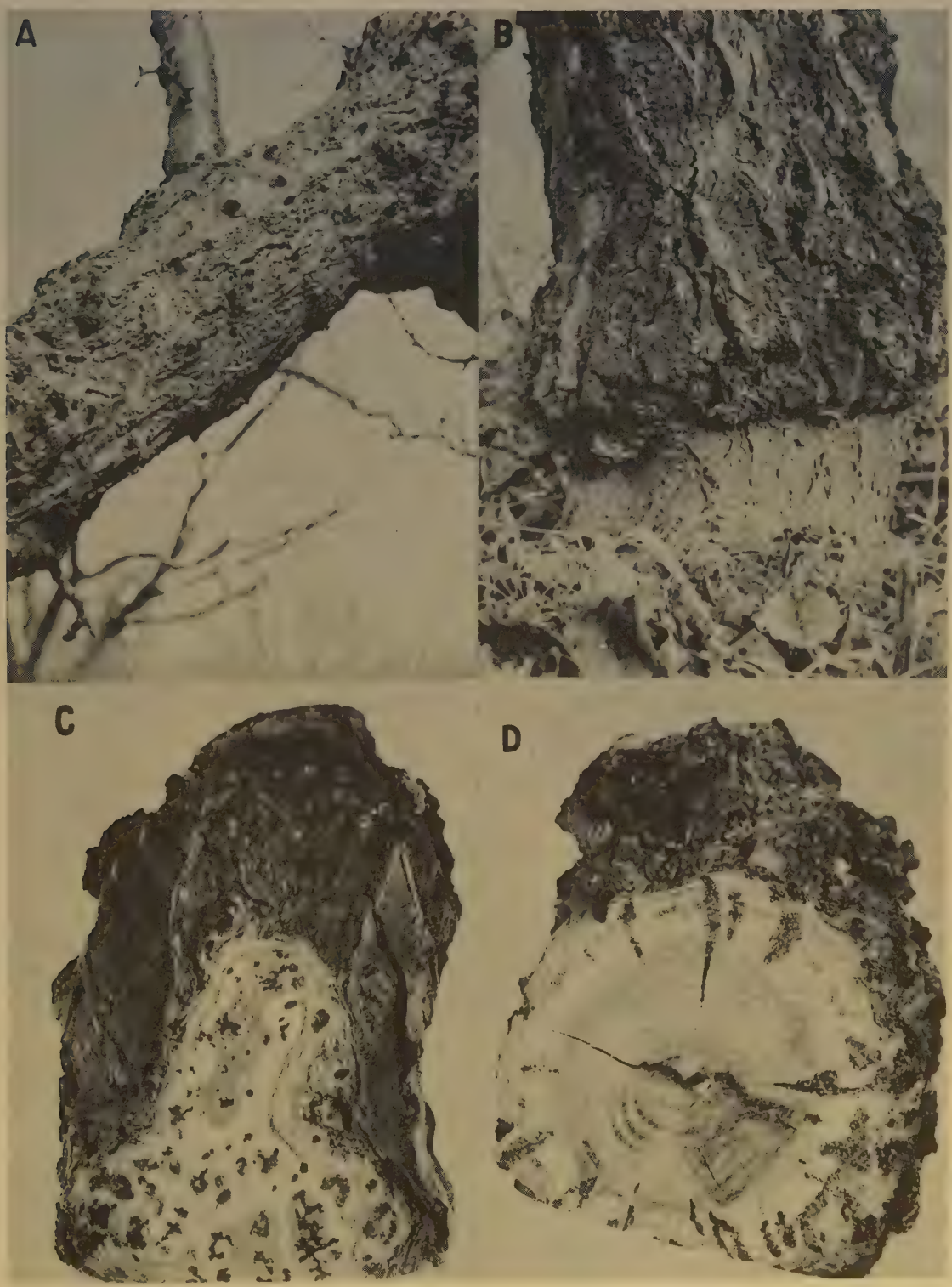

FIGURE 1. Apricot gumboil. A -- Discrete swellings and rough bark of a scaffold limb. B -- Rough bark of the trunk of an affected tree showing overgrowth at the union with the myrobalan understock. C -- Tangential view of an affected branch with the bark and some wood removed to show the gumboils and pockets of necrotic tissue. D -- Cross-section of the same branch as in C showing the occurrence of continuous gum pockets from the inner wood to the surface of the bark.

buds from diseased trees were set in apricot and myrobalan seedlings in the nursery at Davis. The nursery trees were transplanted to a field plot and June-budded above the inoculation buds with healthy Royal and Tilton. The healthy buds were forced, and a few diseased buds also grew into shoots. In 1958 it was observed that only the shoots originating from diseased buds showed symptoms. The gumboil symptoms ended abruptly at the line of union with the apricot seedling stocks. Also in 1953, buds and bark chips from shoots showing symptoms of gumboil were set in 3-year-old Royal and Tilton trees. There has been no evidence of transmission of the gumboil disease in these trees after six growing seasons.

In 1954 seven trees were propagated on Lovell seedlings from one diseased tree to determine whether the gumboil condition was perpetuated in each bud. No evidence of perpetuation has yet been obtained in this test. In transmission tests where perpetuation occurred, the rootstocks were either myrobalan 20 or apricot seedlings. It is possible that the peach rootstock delayed symptom expression. Determining whether delay is consistent and symptom expression is suppressed entirely on peach rootstock will require additional propagation of bud samples from different parts of affected trees.

In 1957 three Royal apricot trees affected with gumboil were grafted with healthy Royal and Tilton scions. Two years later, necrotic gum pockets extended exactly to the line of union of these grafts, and not into the wood of the scions. It is considered too early to draw conclusions from this test of the transmissibility of gumboil.

DISCUSSION

In earlier transmission tests, diseased and healthy scions were grafted on myrobalan 29. Diseased scions were also grafted on apricot seedlings. Transmission would depend on the ability of a virus to move through myrobalan to the healthy scions or into apricot seedlings and express symptoms there. Lack of symptoms in the healthy apricot scions and apricot seedlings does not constitute clear proof of lack of transmissibility. It is possible that if a virus is associated with this disease it cannot move through myrobalan and/or does not show symptoms in certain apricot seedlings. Proof of transmissibility or lack of it must be obtained from experiments in which direct contact occurs between diseased and healthy tissue of the same clone. The evidence at hand does not exclude the possibility that the diseased trees actually are not of the Royal variety; it is possible that all diseased trees originated from a seedling clone in a nursery row of the Royal variety, and that this clone was subsequently propagated along with Royal when budwood was obtained from young orchard trees. Another possibility is that diseased trees had their origin from a somatic mutation of Royal. Experiments in progress should eventually help determine the nature of the gumboil disease of apricot.

Control

The fact that trees propagated from affected trees do not express symptoms for several years after propagation increases the possibility that buds from these trees might be used by nurserymen, thereby spreading the disease in nursery stock. The disease can be avoided if propagators use only healthy, mature trees or trees of known history as sources of propagating stock. Diseased trees can be left in the orchard as long as they remain economically profitable, since there is no evidence of natural spread.

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NOTES ON THE HOSTS OF THREE PINE DWARFMISTLETOES IN NORTHERN COLORADO

Frank G. Hawksworth and Roger S. Peterson[1]

Different species of dwarfmistletoe (Arceuthobium spp.) are typically associated with the three common pines in the northern Front Range of Colorado. Lodgepole pine (Pinus contorta Dougl.) is the usual host of A. americanum Nutt. ex Engelm. Ponderosa pine (P. ponderosa Laws.) is most commonly attacked by A. vaginatum f. cryptopodum (Engelm.) Gill, and limber pine (P. flexilis James) is the usual host of A. campylopodum f. cyanocarpum (Nelson) Gill. These pines typically grow in pure stands, but where they occur in mixtures, the dwarfmistletoe common on one pine may sometimes attack another. All nine combinations between these three pines and the three species of dwarfmistletoe have been found in this area (Table 1). However, the presence of these dwarfmistletoes on trees other than their principal host is of little or no practical importance.

Lodgepole pine is the principal host of A. americanum, but ponderosa pine is also attacked where this tree grows in association with lodgepole pine (2, p. 163, 3). Limber pine has been recorded as a host of A. americanum in Montana (1), but no specimens were preserved and Gill (2) considered the identification of the parasite questionable. A. americanum on limber pine has recently been found in several localities in Boulder and Larimer Counties, Colorado.

A. vaginatum f. cryptopodum is usually associated with ponderosa pine and is rare on lodgepole pine (3) and very rare on limber pine. Only a single limber pine infected by this parasite has been observed; this tree was found about 5 miles north of Nederland, Colorado, along the Nederland-Ward highway.

A. campylopodum f. cyanocarpum is typically associated with limber pine and other five-needled pines. In a stand about 3 miles south of Ward, Colorado, ponderosa pine and lodgepole pine were infected by A. campylopodum f. cyanocarpum where the dwarfmistletoe was

Table 1. Relative abundance of three dwarfmistletoes on three pines in northern Colorado.

Host	Dwarfmistletoe		
	A. americanum	A. vaginatum f. cryptopodum	A. campylopodum f. cyanocarpum
Lodgepole pine	Principal host (Gill 1935, p. 163)	Rare (Hawksworth 1956)	Rare (New record)
Ponderosa pine	Occasional (Gill 1935, p. 163)	Principal host (Gill 1935, p. 179)	Rare (New record)
Limber pine	Rare (New record for central Rocky Mountains)	Rare (New record)	Principal host (Gill 1935, p. 205)

abundant on limber pine. Ponderosa pine seemed to be more susceptible to this parasite than did lodgepole pine. A few ponderosa pines killed by it were seen.

Kuijt (4) suggests that for some forms of A. campylopodum Engelm. in British Columbia there are morphological differences in the plants, depending on the host species. No differences associated with the host were noted in any of the Colorado collections, and the three dwarfmistletoes were readily recognizable regardless of which host they were found on. Figure 1 illustrates an unusual case of parasitism of a ponderosa pine branch by two species of dwarfmistletoe and shows that the morphological characteristics of each species are retained.

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FIGURE 1. A ponderosa pine branch with infections of both Arceuthobium americanum (A) and A. vaginatum f. cryptopodum (V). Pennock Pass, Roosevelt National Forest, Colorado, August 1958.

Shoots on infections of A. americanum and A. vaginatum f. cryptopodum on limber pine were not common and some of these witches' brooms bore no shoots. Weir (5, p. 27) records similar observations on white-barked pines (Pinus albicaulis Engelm.) infected by A. americanum in Montana.

Collections of the host-parasite combinations discussed here have been filed in the herbarium of the Fort Collins Forest Insect and Disease Laboratory.

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EFFICIENT INCORPORATION OF GRANULAR FUNGICIDES AND OTHER CHEMICALS IN THE ROOT ZONE OF CULTIVATED SOILS

A. G. Newhall[1] and W. W. Gunkel

Summary

Tests run on sandy soil at Ithaca to compare the mixing abilities of several tillage implements, as spike tooth harrow, disc, rotary tiller with knives and with spikes gave results strikingly in favor of rotary tools with L-shaped blades.

The past 15 years have seen a remarkable upsurge of interest in the control of diseases and pests attacking the roots of plants. The discovery of good liquid volatile soil fumigants for nematode control was soon followed by the development of ingenious equipment for accurate large-scale application. Unfortunately, cheap volatile liquid fungicides have not been discovered, although progress with insoluble powders has been made. The control of several insect pests with inert granules impregnated with insecticides and the possibility of combining nematocides and fungicides with insecticides in this manner is attracting much deserved attention for several valid reasons. However, this raises the question as to the best methods of incorporating such granules effectively in the top soil and distributing them evenly down to the plow sole, or deeper.

The writer has seen some very disappointing results of root knot control on carrots with powdered and granular nematocides that were incorporated by discing. Many vegetable growers have no tillage tools for land preparation other than a double disc and little is known about the use of the double disc for effectively incorporating granular materials evenly in the soil.

METHODS

After some preliminary laboratory tests with the recovery of sawdust, iron filings, and seeds of different sizes from mineral soil, it was decided to employ seeds of sorghum and of cowpea. These were distributed over the surface of a sandy loam soil previously prepared by plowing, discing, and smoothing with a rake. As an aid to even distribution a 3 x 6 foot wire screen of 1/4-inch mesh was laid on the soil and the seeds were then sown through it (Fig. 1). One and 1/2 pints (550 grams) of cowpea seed or 3/4 pint (200 grams) of sorghum was sufficient for this area, and two or three such areas together constituted one plot for discing and for harrowing, while one was sufficient for rotovating (Fig. 2). In one trial an Ariens and in another a Rototiller was employed, but since both had L-shaped blades and similar speeds the results were very similar. A Merrytiller with straight spike teeth was compared with the rototiller and was much less effective in carrying the seeds down to the lower levels. This is no indictment against the Merrytiller, but merely against the spike tooth form of rotary tiller for this purpose. An 8-foot double disc and similar spring tooth harrow drawn by a Ferguson tractor were compared on a sandy loam soil. The disc was found superior, as Figure 3 indicates, but was still not good enough.

RESULTS

Results are expressed as percentages of the total seeds recovered from each of three levels (Fig. 3). The thickness of each layer was that of a greenhouse flat or tray, namely, 2 3/4 inches. Seeds were screened from this soil and counted. This procedure gives little indication of lateral distribution, but since the plow was not used in these tests and it was mainly vertical distribution that was wanted, this fact was ignored. It is quite evident that the only satisfactory tool was the rotary tiller with bent knives, and that two passes, back and forth, are better than one. The spike tooth rotary tool was very ineffective even with repeated passes (up to five) and so was the spring tooth harrow four times across the field, set at maximum possible depth. The disc, likewise, failed to bury particles to sufficient depth even with four passes, back and forth, during which operation, it might be recorded, some seeds were

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[1] Acknowledgment of the help of Wilfred Arnold is gladly made here.

FIGURE 1. A 3 x 6 foot screen of 1/4-inch mesh facilitated the even distribution of the same number of seeds per unit area.

FIGURE 2. Replicated plots 3 x 6 feet were ample for rotary tilling, which was done to maximum depth of 8 inches.

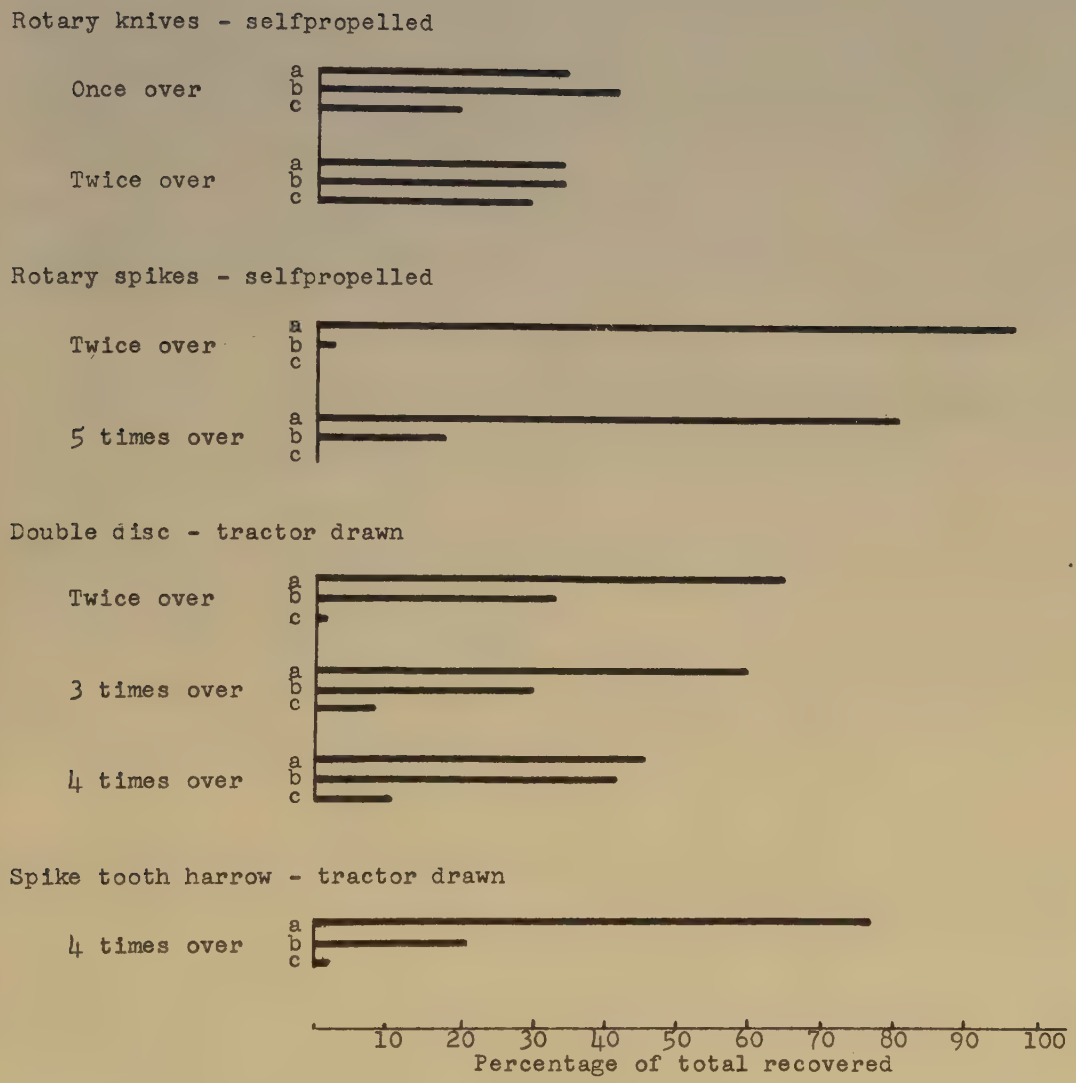

FIGURE 3. Vertical distribution of adjuvants in top soil tilled with different tools.

carried 10 to 12 feet out of the plot in each direction.

The conclusion is reached that if an even distribution of chemicals clear to the plow sole is needed for nematode or root rot control, then, we should be thinking more in terms of some efficient rotary tool for their incorporation. Just who is to furnish the grower with such equipment is the $64 question, but it is becoming clearer that unless the grower gets results with granular materials he cannot be expected to come back for more. This paper merely points out one reason why the grower may not be getting results. The conclusions drawn are essentially similar to those of Hulburt and Menzel[2] in their more extensive study regarding the relative effectiveness of ordinary farm tools for these purposes. Nearly all cultivated soils contain lumps, no matter how carefully the soil is prepared. Some penetration of these lumps can take place with volatile toxicants and with soluble ones if the soil is drenched. However, with relatively insoluble toxicants, our observations indicate that very little, if any, penetration of lumps ever takes place with ordinary tillage operations. It is admitted that for some kinds of insect and weed control it may not be necessary to incorporate chemicals as deeply as it would be for nematodes and root rot. Let us not blame materials until we know they have been adequately distributed where needed.

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[2]Hulburt, W. C., and R. G. Menzel. 1953. Soil mixing characteristics of tillage implements. Agricultural Engineering. 34: 702-708.

A TECHNIQUE FOR ASSESSING LEAF COVERAGE WITH ZINC FUNGICIDES[1]

H. R. MacCarthy[2]

Common zinc fungicides were used to demonstrate spray coverage in potato crops. A fairly quick and simple procedure was originated by Blodgett and Mader in 1934 (1) for printing copper spray patterns from leaves on to paper. The idea was developed and simplified to give reproducible results with the use of a sensitive zinc indicator.

MATERIALS AND METHODS

Dithizone (diphenylthiocarbazone) was used as the indicator (2). This compound forms insoluble purple-red complex salts with zinc in neutral, alkaline and acid solutions. The following proportions were used:

100 mg dithizone
75 ml carbon tetrachloride
200 ml 95 percent ethyl alcohol

The dithizone was dissolved in the carbon tetrachloride and the alcohol added. This gave a dark green solution. The proportions were not, apparently, critical. The solution was prepared and stored in Pyrex glass containers in order to avoid contamination with heavy metals.

Single sheets of Whatman No. 1 filter paper of appropriate size were dipped with forceps into a shallow dish of the solution. The surplus was allowed to run off, and the sheets were air dried without touching metal or one another. Wooden clothes pins strung on a cord were used for drying. The sheets were stored in the dark in Pyrex glass. Their effective life is not known. Large potato leaflets fitted easily on 11.0 cm circles.

In use, a leaf or leaflet was placed between two sheets of sensitized paper. A stack of such pairs, each pair separated from the next by a folded paper towel, was put in a laboratory press. The pressures used were about 5000 pounds per square inch for 10 seconds but such high pressure is probably not necessary so long as the leaf is flattened.

RESULTS

Potato leaflets partly dipped in a test solution of 500 ppm of zinc sulfate plus spreader and then dried, left a bright purple-red. (Fig. 1, 1A, 1B). In testing potato leaves power sprayed with zineb in the field 2 or 3 days before at fairly low gallonage (40 imperial gallons per acre) and normal concentration (3 pounds per acre) the impression was pink and somewhat less bright but was easily read. Potato leaves hand sprayed with zineb at normal rates left a clear impression when printed 2 days later (Fig. 1, 3A, 3B). Zineb (Fig. 1, 5A, 5B) and ziram dusts on pole beans were as easy to read as were sprays. The additional coverage resulting from the use of a spreader in the spray tank was clearly demonstrated.

The impressions were difficult to photograph in black and white, especially those of the check leaves (Fig. 1, 2A, 2B, 4A, 4B), which were pale green, slightly darker along the veins. In impressions of treated leaves the color contrast is striking and the pink areas are instantly and clearly recognizable.

ACKNOWLEDGMENTS

Thanks are due to Dr. P. M. Townsley and Mr. D. L. Hatt, Canada Department of Agriculture, Vancouver, for advice on the choice of an indicator.

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[1]Contribution No. 9, Science Service Laboratory, Canada Department of Agriculture, c/o University of British Columbia, Vancouver, B. C.
[2]Associate Entomologist.

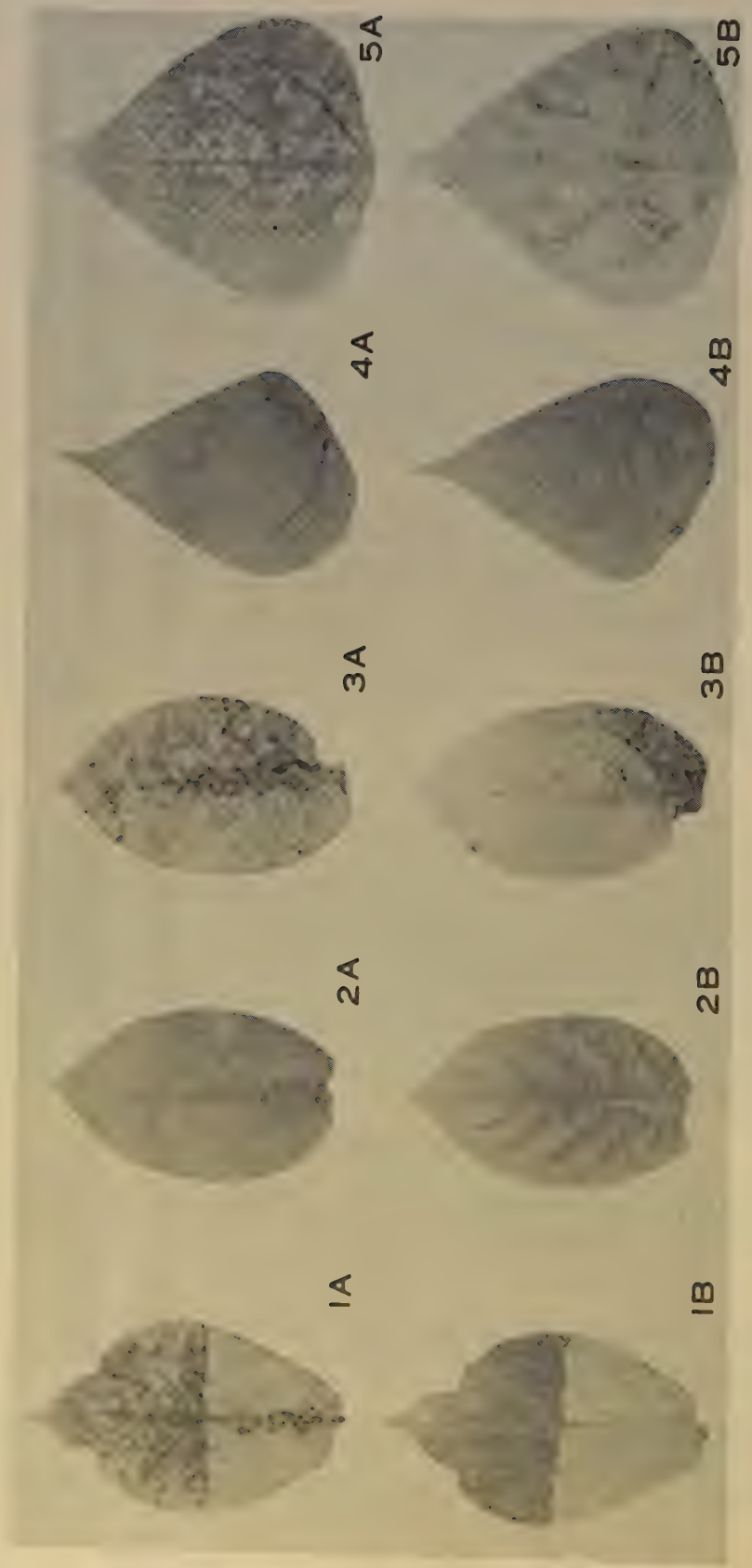

FIGURE 1. A and B are impressions of upper and lower surfaces respectively. 1A, 1B, potato leaflet, partly dipped in solution of 500 ppm zinc sulfate with spreader; 2A, 2B, potato leaflet untreated; 3A, 3B, potato leaflet, hand sprayed in field with zineb; 4A, 4B, pole bean leaflet untreated, 5A, 5B, pole bean leaflet dusted twice in field with zineb, 7 and 23 days previously.

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BRIEF NOTES

CHLOROSIS OF SPINACH ASSOCIATED WITH OLPIDIUM BRASSICAE

By Saul Rich

This year the spinach crop in Connecticut was badly damaged by a chlorosis of the older leaves. Often the tissue at the center of the chlorotic area was collapsed. The disease appeared after a few warm, sunny days following a prolonged wet, cool period, and was most severe in wet, poorly drained areas of the field. The symptoms appeared on plants growing in both light, sandy soils and in heavy, clay soils, and in soils varying in pH from 5.5 to 6.5. Spinach in the heavier soils was more severely damaged than spinach in light soils. The plants did not respond to foliar sprays of either manganous sulfate or magnesium sulfate.

The roots of the chlorotic plants were very poor and heavily infected with Olpidium brassicae. Infection studies are now in progress to determine if the Olpidium is responsible for the poor root condition and leaf chlorosis.

On one farm, lettuce in a field adjacent to the spinach also was infected with Olpidium and had big vein[1].

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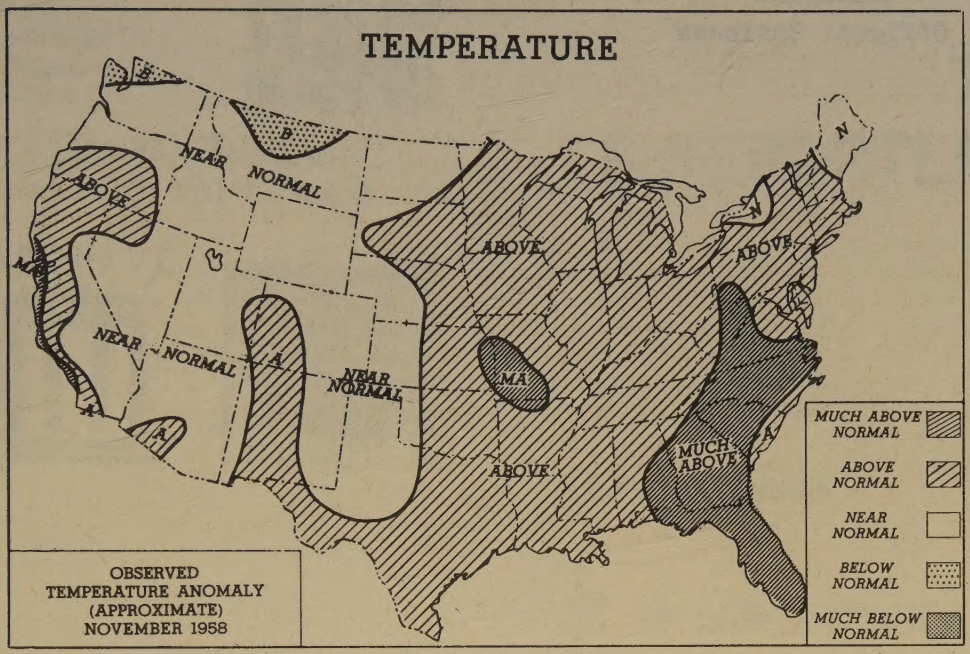
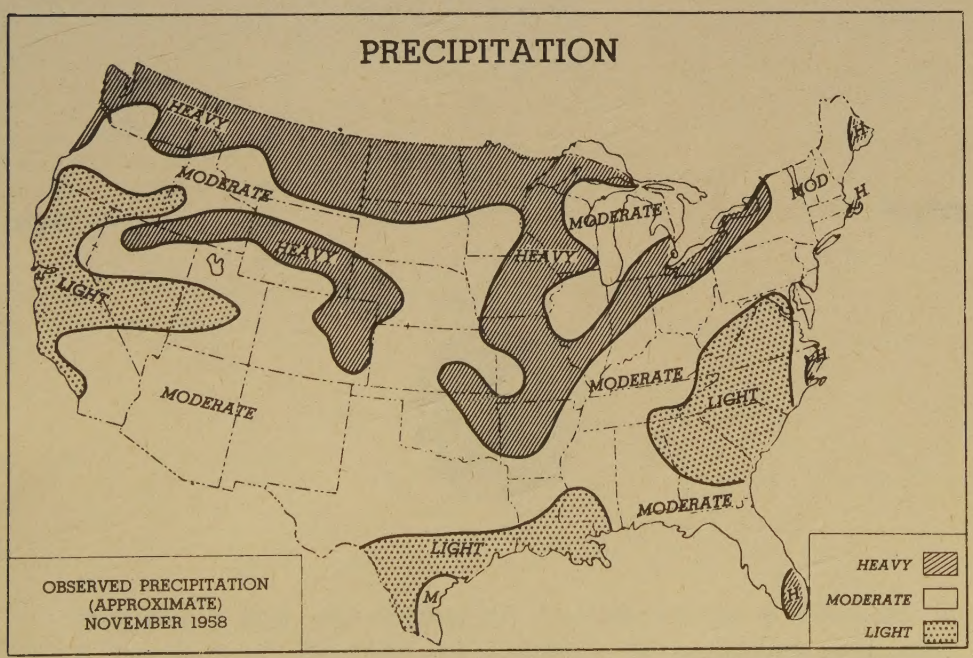
[1] Grogan, R. G., F. W. Zink, W. B. Hewitt, and K. A. Kimble. 1958. The association of Olpidium with the big vein disease of lettuce. Phytopathology 48: 292-297.

CORRECTION

REPORTER, October issue (Volume 42, Number 10), page 1170: In the first paragraph, change line 4 to read "... planting test and check plants on opposite sides of the same furrow. In either case, root systems ..."





The terms used in the accompanying maps, which define the ranges of temperature and precipitation, are numerical class limits. These are based on a statistical analysis of past records through which is determined the normal frequency of occurrence of temperatures and precipitation at various times of the year for different locations. For temperature the classes above, below, and near normal are so defined that they each normally occur one-fourth of the time; much above and much below normal, one-eighth of the time. Precipitation is depicted in terms of light, moderate, and heavy, each class normally occurring one-third of the time and thereby having equal probability of occurrence. These maps graphically represent only the general trends and give the country's weather picture at a glance. For quantitative studies, where monthly mean temperatures and actual precipitation records are needed for a given time and place, other publications of the Weather Bureau should be consulted. P. R. M.

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